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***Pelagic Organism Decline (POD):
Acute and Chronic Invertebrate and Fish Toxicity
Testing in the Sacramento-San Joaquin Delta
2006-2007***

Final Report

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1. Executive Summary

This report covers the project period of January 1, 2006 – December 31, 2007. Relevant information obtained in 2005 is also included. The study described here encompasses a sampling and toxicity monitoring program in the Sacramento-San Joaquin Delta (including several sites in Suisun Bay and the Napa River), and the development of molecular stress indicators for two fish species of concern, delta smelt (*Hypomesus transpacificus*) and striped bass (*Morone saxatilis*). Biweekly toxicity tests were performed using *Hyaella azteca*, an amphipod species resident in the Delta. Toxicity testing protocols were developed for larval and juvenile delta smelt and juvenile striped bass, and water samples from the Delta were tested during late spring/early summer using these fish species.

Water Quality at Field Sites: Site-specific water quality parameters were routinely monitored in the field at the time of sampling. During the project period, temperature ranged from 5.8 (site 902) to 28.6°C (Light 55), dissolved oxygen from 5.1 (site 609) to 13.9 mg/L (Light 55), specific conductivity from 86 (site 915) to 30,260 µS/cm (site 323), pH from 7.6 (Hood) to 8.7 (site 915) and turbidity from 1.4 (site 504) to 219.7 NTU (site 323). Mean total ammonia-N concentrations were highest at stations Hood and 711, both on the lower Sacramento River however, annual and seasonal differences were apparent. Unionized ammonia concentrations were highest at sites 405 (Benicia), 711 (Sacramento River nr. Rio Vista) and Light 55 (Sacramento Deep Water Channel). Other sites with seasonally high NH₃ concentrations were 902 (summer 2006), 602 (winter 2007), and 910 (spring 2007).

Toxicity Monitoring with *H. azteca*: Monitoring sites were selected among the California Department of Fish and Game Towntnet Survey stations, and in accordance with the prevalent distribution patterns of fish species of concern. Water samples were collected twice a month at sites 323, 340, 405, 504, 508, 602, 609, 704, 711, 804, 812, 902, 910, 915, and Light 55 in the Sacramento River Deep Water Channel (for more detailed information see Table 1), and tested using a 10-day *H. azteca* bioassay with growth and survival as chronic and acute endpoints, respectively. Routine partial toxicity identification evaluation (TIE) tests were conducted on all water samples with the chemical piperonyl-butoxide (PBO), a chemical synergist/antagonist, to provide early evidence for the presence of two classes of toxic insecticides, organophosphates and pyrethroids. If toxicity (>50% survival within 7 days) was observed in a water sample, TIEs were initiated immediately to identify the causative agents. Water samples were submitted for chemical analyses whenever significant acute or chronic toxicity was observed.

Acute Toxicity to *H. azteca*: Of 693 water samples tested during the project period, fifteen (2.2%) caused a significant reduction in amphipod survival. Most of these were collected from sites in the lower Sacramento River (Hood, site 711), the Deep Water Shipping Channel (Light 55) and site 405 (Benicia). In addition, one sample collected on 7/10/07 from site 602 (Suisun Bay) and one sample from site 323 (7/12/06, San Pablo Bay) were acutely toxic. The majority of toxic samples (93.3%) were collected in 2007, mostly during the second half of the year (July-December). Samples from the Sacramento River at Hood were only tested in the second part of 2007, and 38% of these samples were acutely toxic to *H. azteca*. The observed pattern suggests an inverse

relationship of toxicity with river flows, and compromised water quality in the lower Sacramento River/Deep Water Shipping Channel and Carquinez Strait near Benicia.

PBO Effect on H. azteca Survival: Significant changes in acute toxicity due to addition of PBO were seen in seven samples (1%) collected from the Sacramento River at Hood, sites 711, 704, Light 55, 340 (Napa River), 405 (Benicia) and 323 (San Pablo Bay). The observed response suggests the presence of organophosphate insecticides in samples collected from Hood and Light 55 (both in Oct. 2007), and the presence of pyrethroid insecticides in samples collected at site 323, 340, and 405. There is some evidence that pyrethroids were also present in samples collected on July 10, 2007 from sites 804, 504 and 508, three adjacent field sites. TIEs performed on toxic samples from sites 323 and 405 indicated that non-polar organic chemicals contributed to the observed toxic effects, while toxicity was lost in samples from sites 711 and Hood by the time TIEs could be performed (ca. 14 days after sample collection). Analytical chemistry confirmed that the sample from site 340 contained 3 ng/L cyfluthrin and 16 ng/L esfenvalerate, and two samples from site 405 contained 3 ng/L esfenvalerate, and 5 ng/L permethrin, respectively, but most samples did not contain detectable amounts of insecticides. Studies to trace the fate of pyrethroid insecticides during sampling and testing are scheduled.

Chronic Toxicity to H. azteca: Addition of PBO to the ambient sample resulted in a significant reduction or increase in amphipod growth (relative weight at test termination) when compared to the ambient sample in a total of 33 water samples (4.9% of samples tested). PBO addition led to increased growth in 3, and decreased growth in 30 samples. Significant PBO effects were detected in 14 samples in 2006 (4.1%), and 19 samples in 2007 (5.7%). Water samples where PBO addition resulted in a reduction in growth were primarily collected from sites in the South-Eastern Delta (902, 910, 915), the lower Sacramento River (Light 55, 711) and Suisun Bay (609, 602, 508). Three water samples where PBO addition resulted in an increase in growth were collected from sites 902, 910 and 812 on June 6, 2007. Patterns where several neighboring sites sampled on the same date triggered the same response in bioassay organisms were seen repeatedly, and most of these samples were collected in the spring or summer. Several of these samples contained detectable amounts of pyrethroid pesticides: Site 902 sampled on 8/22/06 contained 5 ng/L cyfluthrin and 24 ng/L permethrin; site 340 sampled 2/13/07 contained 63 ng/L cyfluthrin, and sites 915 and 508 sampled on 2/28/07 and 3/1/07, respectively, contained 2 and 3 ng/L lambda-cyhalothrin. A sample from Light 55 collected 2/1/07 contained 6 ng/L diazinon.

Site-Specific Growth of H. azteca: Growth data from toxicity tests conducted during 2006 with *H. azteca* was analyzed to determine if any site-specific effects on growth were detectable. No strong evidence was found that would suggest major site-to-site or seasonal differences in *H. azteca* growth that could not be accounted for by differences in conductivity. However, season-specific analysis of growth data revealed trends in growth deviations from expected values at sites 711, 405 and 602 (lower) and 704, 804, 902, 915 (higher).

Effect of Ammonia on H. azteca Survival and Growth: Analysis of the entire dataset revealed that ammonia-N and unionized ammonia had significant effects on *H.*

azteca growth, but no significant effect on *H. azteca* survival. When analyzed by site, total ammonia-N concentrations were negatively correlated to survival at Light 55, but positively correlated to amphipod survival at sites 504, 609 and 804. Ammonia-N and unionized ammonia concentrations were negatively related to *H. azteca* growth at sites 323, 812 and Light 55. Analysis of ammonia effects across sites for different seasons determined that survival and growth during the winter of 2007 was negatively associated with levels of ammonia-N and unionized ammonia.

Laboratory Experiments with *H. azteca*: A study was performed to evaluate the toxicity of environmentally relevant concentrations and mixtures of two pyrethroid pesticides detected in a water sample collected on August 22, 2006 at Site 902 (Old River at the mouth of Holland Cut. The LC50 for cyfluthrin alone was determined to be 0.0065 ppb, and the LC50 for permethrin alone was estimated to be 0.0465 ppb. The addition of 25 ppb PBO doubled the toxicity of cyfluthrin and more than tripled the toxicity of permethrin. The permethrin and cyfluthrin mixture resulted in higher, but slightly less-than-additive toxicity than either pesticide alone.

Toxicity Monitoring with Striped Bass: To date, just a few pilot tests have been performed with larval striped bass due to the difficulties in obtaining larvae of this particular strain of striped bass. Two tests with juvenile (80-90 d old) fish were conducted with water collected from sites 340, 508, 609, 711, 910 and 915 on July 30, 2005 and August 25, 2006. No significant effects on survival or growth were observed. The sensitivity of juvenile (81-90 d) striped bass to two individual toxicants, copper and the pyrethroid insecticide esfenvalerate was investigated. The 7-d LC50 for copper was determined to be 254 $\mu\text{g/L}$ Cu^{2+} (dissolved). For esfenvalerate, the 24-h LC50 was 2.17 $\mu\text{g/L}$, and the 24-h EC25 (swimming behavior) was 1.07 $\mu\text{g/L}$.

Toxicity Monitoring with Delta Smelt: Test protocols were developed for toxicity tests using delta smelt larvae at different stages of development (20-92 d) and juveniles. While static renewal tests were performed in 2006, a flow-through system was used in 2007. This system proved to be superior to the static renewal method. Delta smelt were obtained from the UC Davis Fish Conservation and Culture Laboratory, Byron, CA, and exposed for 7 days to water samples from sites 711, 910, 915, 609, 504 and 340 (2006), or 711, Hood, 915, Vernalis, 609, 504 and 340 (2007) as well as EC and turbidity controls. The sensitivity of delta smelt to copper and the pyrethroid insecticide esfenvalerate, was investigated.

Turbidity and EC/salinity were the two most important factors determining survival of delta smelt larvae overall, particularly for larvae less than 44 days old. These younger larvae (20-36 d old) tended to survive poorly in low EC samples from the lower Sacramento River, Old River and the San Joaquin River, as well as in the low EC control (150-180 uS/cm) even when turbidity was adjusted to 10 NTU. Their survival was highest in water from the Napa River (site 340), and Montezuma Slough (site 609), which had both saline ($\text{EC} > 4000$ uS/cm) and the most turbid water. Larvae that were 44 d old and older appeared to be less dependent on high turbidity and salinity. Survival was significantly lower than in the low EC control among delta smelt exposed to samples from Hood (collected June 6, 2007) and site 711 (July 26, 2007), both in the lower Sacramento River. Although EC and turbidity were low at these sites, the reduced survival cannot be explained by these factors alone.

Effect of Ammonia on Delta Smelt Larvae: Data analysis results showed a significant correlation of unionized ammonia concentrations (maximum laboratory value measured during test) and larval survival in static renewal tests performed in 2006, with an approximate LC50 of 0.012 mg/L NH₃. The same analysis on 2007 data showed no such correlation. Taking into account the effects of EC, statistical analysis of the complete 2006-2007 data showed no correlation of larval 7-d survival with NH₃ concentration in our tests, where maximum unionized ammonia concentrations were <0.016 mg/L. However, in the 2006 data set we continued to see a marginally significant (p=0.06) correlation of 7-d survival and unionized ammonia. It is important to note that the laboratory tests were carried out with larvae of different ages (20-92 days old). Targeted tests to determine ammonia toxicity to delta smelt are scheduled.

Reference Toxicants: A 7-day test with juvenile (90 d) delta smelt yielded LC50 values for copper toxicity of 33.5 µg/L (96 h), and 24.7 µg/L (7 d). The 24-h LC50 of the pyrethroid esfenvalerate for 10-d to 204-d old delta smelt was 0.1-0.76 µg/L (nominal conc.), and the 24-hEC25 for swimming impairment was 0.03-0.28 µg/L, indicating that delta smelt larvae are highly sensitive to this insecticide, and that sensitivity is inversely related to age/size.

Sublethal Indicators of Contaminant Effects in Delta Species: In an effort to develop field-applicable tools for the detection of stressor-specific, sublethal effects in striped bass and delta smelt tissues, biochemical and molecular biomarker protocols were developed and applied.

Inhibition of Acetyl-Cholinesterase in Brain and Muscle Tissue of Juvenile Striped Bass and Delta Smelt: For organophosphate (OP) and carbamate insecticides, the primary mechanism of toxic action is the inhibition of the enzyme acetylcholinesterase (AChE), which is commonly used as a diagnostic tool for sublethal OP and carbamate exposure and effect. For this study, we quantified AChE activity in brain and muscle of juvenile striped bass and delta smelt exposed to water samples from the Delta or to copper. No effects on AChE activity were seen after exposure to water samples from the Delta collected on July 27/28, 2005 (striped bass) and on August 30/31, 2005 (delta smelt) from CDFG stations 340, 711, 910 and 915. Copper did not affect AChE activity at sublethal Cu⁺ concentrations however, at 50 ppb Cu⁺ enzyme activity in the brain of delta smelt was significantly reduced.

Expression of Stress-Responsive Genes in Striped Bass Exposed to Copper and Esfenvalerate: Method development and results of laboratory tests were published by Geist et al. (2007). The effects of two reference toxicants, copper (Cu) and the pyrethroid insecticide esfenvalerate, on lethal (mortality) and sublethal endpoints (growth, swimming behavior, and transcription levels of stress response genes) were investigated in juvenile (81-90 d old) striped bass (*Morone saxatilis*). Cellular stress response markers for proteotoxicity (HSP70, HSP90), phase I detoxification mechanism (CYP1A1), metal-binding (metallothionein), as well as immune-function and pathogen-defense (TGF-β, Mx-protein, nRAMP) were developed. Quantitative real-time TaqMan-PCR was used to examine tissue-specific changes in the transcriptome of liver, spleen, white muscle, anterior kidney and gills after 7-d Cu exposures and 24-h esfenvalerate exposures. On the transcriptome level, exposure to Cu showed strongest effects on the transcription of

metallothionein in spleen tissue, causing a 4-fold increase of mRNA at 42 ppb total Cu and a 10-fold increase at 160 ppb Cu. Exposure to Cu also caused significant tissue-specific changes in gene transcription for immune-system related genes. Esfenvalerate exposure had tissue-specific effects on the transcription of HSP70, HSP90 and CYP1A1. The most significant effects were detected in liver tissue after exposure to 0.64 µg/L esfenvalerate. Results show that the stress response at the transcriptome level is a more sensitive indicator for Cu and esfenvalerate exposures at low concentrations than swimming behavior, growth or mortality.

Expression of Stress-Responsive Genes in Striped Bass Exposed to Delta Water Samples: Tissue samples of juvenile striped bass exposed for 7 d to Delta water samples collected on August 22/23, 2006 from CDFG stations 340, 508, 609, 711, 910 and 915, were analyzed for the following stress- or contaminant-responsive genes: for proteotoxicity (HSP70, HSP90), phase I detoxification mechanism (CYP1A1), metal-binding (metallothionein), immune-function and pathogen-defense (TGF-B, Mx-protein, nRAMP) as well as estrogenic endocrine disruption (Vitellogenin). Significant responses were seen at sites 910, 609 and 711. Data analysis is ongoing.

Expression of Stress-Responsive Genes in Striped Bass Exposed to SPMD Extracts from Delta Sites: To assess the presence and effects of bioavailable lipophilic contaminants in the estuary Semi-Permeable Membrane Devices (SPMDs) were deployed in three locations in the Delta from August 16 to September 13, 2005, by D. Ostrach, UC Davis. SPMDs bind nonionic organic compounds and some neutral organo-metal complexes. SPMD extracts were used in 3-d injection experiments with striped bass. Spleen and liver samples were analyzed for molecular biomarkers described above. SPMD extracts from all three field sites produced gene responses in the liver, but not the spleen, of exposed fish. Extract from the Collinsville site down-regulated transcription of Cyp1a1 and Mt, while extracts from Sand Mound and Napa down-regulated transcription of Mt only. Vitellogenin was slightly increased in fish exposed to SPMD extracts from Collinsville. Further data analysis is ongoing.

Expression of Stress-Responsive Genes in Delta Smelt (DNA-Microarray): In order to understand the effects of contaminants upon *Hypomesus transpacificus* a microarray with over 8,000 Expressed Sequence Tags (ESTs) was constructed and applied to measure gene responses on 60-day old juveniles exposed to 50 µg/L copper for 7 days. The sublethal effects of copper exposure in the delta smelt appear to be on neuro-muscular activity, respiration and metabolism. Expression of a number of genes involved in cardio-muscular contraction, neuro-transmission, oxidative stress, metal ion binding, immunity and systemic inflammation, and digestion was altered in response to copper exposure. Amongst the responding genes there was a significant up-regulation of osteonectin, a source of copper-binding peptides, which may be indicative of tissue damage caused by excess copper. Future work will include additional microarray analyses of delta smelt exposed to different toxicants, and investigation of a selected suite of genes from these microarray assessments, using real-time quantitative PCR to develop informative molecular biomarkers of stress and exposure in the delta smelt.

2. Background and Approach

In the last several years, abundance indices of numerous pelagic fish species residing in the Sacramento-San Joaquin Delta of California, USA, have shown marked declines and record lows for the endemic delta smelt (*Hypomesus transpacificus*), age-0 striped bass (*Morone saxatilis*), longfin smelt (*Spirinchus thaleichthys*) and threadfin shad (*Dorosoma petenense*) (Stevens and Miller, 1983; Stevens et al., 1985; Moyle et al., 1992; Moyle and Williams, 1990). While several of these species - including in particular longfin smelt and juvenile striped bass - have shown evidence of long-term declines, there appears to have been a precipitous “step-change” to very low abundance during the period 2002-2004 (Bryant and Souza, 2004; Hieb et al., 2005; Feyrer et al., 2007). It is presently unclear what might have caused this critical population decline, but toxic contaminants may be one of several factors acting individually or in concert to lower pelagic productivity.

Agricultural, industrial, urban and mining sources release contaminants into waterways, and water quality assessment studies indicate that the criteria for the protection of freshwater aquatic life have been exceeded in many Central Valley streams (Domagalski et al., 2000; Dubrovsky et al., 1998; DeVlaming et al., 2000; Werner et al., 2000). While measured concentrations of chemical contaminants were generally below acutely toxic levels for fish, sublethal toxic effects may result in energy reallocation, increased susceptibility to disease and predation, reduced reproductive success and behavioral abnormalities, with the potential to decrease evolutionary fitness (Scholz et al., 2000; Sorensen, 1991; DeVlaming et al., 2000; Sandahl et al., 2005, Clifford et al., 2005, Floyd et al., 2008).

Ecological effects of aquatic contaminants are difficult to detect and quantify. Available ecotoxicological tools for screening contaminant exposures in the field include bioassays, Toxicity Identification Evaluation methods (TIEs) or risk assessments based on existing data (Rand, 1995; US EPA, 1989 a, b; 1991; 2000). On a level of higher resolution, altered cellular and molecular responses to stressors can be used as powerful tools for gaining a better understanding of the mechanisms involved, and thus as biomarkers for the identification of environmental impacts on aquatic ecosystems (Huggett et al., 1992). The rising field of ecotoxicogenomics links the two disciplines genomics and ecotoxicology, mostly by identifying cellular biomarkers and biosignals at the transcriptome level as indicators for the exposure to contaminants. In a first step, microarray approaches are used to initially identify suites of up- or downregulated genes, and changes in gene expression of selected genes are quantified subsequently by quantitative real-time PCR. However, for non-model species the high number of unidentifiable genes from random libraries and the comparatively high costs of microarray development and use can pose substantial limitations to this approach. In addition, only few studies simultaneously consider multiple tissues and tissue-specific effects when carrying out studies on the transcriptome.

During a 2005 four-month pilot study involving toxicity testing of Delta water samples, significant acute and chronic toxicity to amphipods (*Hyalella azteca*) was detected at five out of ten sampling sites: the Napa River (340), the Old River (902), the San Joaquin River (910), the San Joaquin River, 1 km upstream from the mouth (804), and the Sacramento River (711) in 6 of

131 water samples tested (4.6%). Our 2006-07 study continued this approach with a spatially and temporally expanded sampling and toxicity testing program. Fifteen sites were sampled twice a month in accordance with the prevalent distribution patterns of fish species of concern. The amphipod species, *H. azteca*, an important component of the Delta ecosystem, was used for routine toxicity testing. This species is resident in the Delta, sensitive to contaminants, and is routinely used in toxicity testing programs throughout the Nation. Routine partial TIE tests (addition of PBO) were conducted to provide early evidence for the presence of two classes of toxic insecticides, organophosphates and pyrethroids. If toxicity was observed at a site through initial screening, Toxicity Identification Evaluation (TIE) procedures were to be initiated immediately to identify the causative agents. In addition to the conventional bioassay approach, molecular biomarkers are being developed and validated for two fish species of concern, striped bass (*M. saxatilis*) and delta smelt (*H. transpacificus*). The expression of certain genes in response to environmental stressors is considered to be more sensitive, and potentially stressor-specific, and is of promise for the identification of stressor impacts in the field.

Questions addressed:

- 1) Is water in the Delta and the Napa River toxic to pelagic fish and fish food organisms?
- 2) What is the spatial and temporal distribution of water column toxicity in areas of the Delta that are important for fish species of concern?
- 3) What are the primary toxicants in Delta water samples?

3. Toxicity Monitoring

3.1 Sampling Sites

Sampling occurred on a bi-weekly basis from the period of 1 January, 2006 through 31 December, 2007 (Tables 1, 2, Fig. 1). Of the 17 sampling sites, six (2006) to seven (2007) were tested with a fish species, delta smelt and striped bass, in addition to invertebrates. Due to a change in testing methods for delta smelt in 2007 from static renewal methods to flow-through exposures, water from the San Joaquin River was collected by car at the DWR Monitoring Station at Vernalis replacing site 910 (sampled by boat in 2006). The DWR Monitoring Station at Hood was added as a sampling site for delta smelt testing in 2007, and we continued testing samples from this site with *H. azteca* from the summer of 2007 until the end of the project period. All sampling sites lie within the greater Sacramento-San Joaquin Delta. Only one sample was collected from "Stockton Port" following a possible fish kill in the vicinity of this site, and tested using fathead minnow larvae and *H. azteca*. In addition, 10 water and 10 sediment samples were collected on 13-15 June, 2006 for chemical analysis of pesticides by the Department of Pesticide Regulation.

Table 1. Sampling stations and GPS coordinates during the 2006-2007 project period.

STATION	LOCATION	Latitude	Longitude
323	San Pablo Bay, Rodeo Flats opposite end of rock wall.	38-02'-53.9"N	122-16'-58.1"W
340	Napa River along Vallejo seawall and park.	38-05'-51"N	122-15'-43.9"W
405	Carquinez Straight, just west of Benicia army dock.	38-02'-22.9"N	122-09'-01.8"W
504	Suisun Bay, east of middle point.	38-03'-16.2"N	121-59'-22.2"W
508	Suisun Bay, off Chipps Island, opposite Sacramento North ferry slip.	38-02'-43.8"N	121-55'-07.7"W
602	Grizzly Bay, northeast of Suisun Slough at Dolphin.	38-06'-50.4"N	122-02'-46.3"W
609	Montezuma Slough at Nurse Slough.	38-10'-01.9"N	121-56'-16.8"W
704	Sacramento River, north side across from Sherman Lake.	38-04'-09"N	121-46'-31"W
711	Sacramento.River at the tip of Grand Island.	38-10'43.7"N	121-39'-55.1"W
804	Middle of Broad Slough, west end.	38-01'-05.5"N	121-47'-49.2"W
812	San Joaquin River, just west of Oulton Point.	38-05'-25.1"N	121-38'-25.8"W
902	Old River at mouth of Holland Cut.	38-01'-09.1"N	121-34'-55.9"W
910	San Joaquin River, between Hog and Turner Cut.	38-0'-06.5"N	121-26'-55.3"W
915	Old River-Western arm at railroad bridge.	37-56'-33"N	121-33'-48.6"W
Light 55	Sacramento River Deep Water Channel at Light 55	38-16'-26.5"N	121-39'-42.9"W
Hood	DWR Water Quality Monitoring Station	38-22'-03.6"N	121-31'-13.6"W
Stockton Port	Downstream of Stockton Waste Water Treatment Plant	37-56'-05.7"N	121-19'-48.2"W
Vernalis	DWR Water Quality Monitoring Station, San Joaquin River	37-40'-45.8"N	121-31'-13.6"W

Table 2. Sampling Dates and Toxicity Tests Performed

Site	Date									
	01/12/06	01/24/06 - 01/25/06	02/07/06 - 02/08/06	02/21/06 - 02/22/06	03/07/06 - 03/08/07	03/20/06 - 03/21/06	04/03/06 - 04/05/06	04/17/06 - 04/18/06	05/01/06 - 05/03/06	05/15/06 - 05/17/06
323	-	H	H	H	H	H	H	H	H	H
340	-	H	H	H	H	H	S/H	H	S/H	S/H
405		H	H	H	H	H	H	H	H	H
504	H	H	H	H	H	H	H	H	H	H
508	H	-	H	H	H	H	S/H	H	S/H	S/H
602	-	H	H	H	H	H	H	H	H	H
609	H	H	H	H	H	H	S/H	H	S/H	S/H
704	H	H	H	H	H	H	H	H	H	H
711	H	H	H	H	H	H	S/H	H	S/H	S/H
804	H	H	H	H	H	H	H	H	H	H
812			H	H	H	H	H	H	H	H
902	H	H	H	H	H	H	H	H	H	H
910	H	H	H	H	H	H	S/H	H	S/H	S/H
915	H	H	H	H	H	H	S/H	H	S/H	S/H
Light 55	-	-	H	H	H	H	H	H	H	H
Vernalis	-	-	-	-	-	-	-	-	-	-
Hood	-	-	-	-	-	-	-	-	-	-
Stockton Port	-	-	-	-	-	-	-	-	-	-

H=Hyalella azteca
 S=Delta smelt
 B=Striped bass
 F=Fathead minnow

Table 2, continued: Sampling Dates and Toxicity Tests Performed

Site	Date									
	05/30/06 - 06/01/06	06/13/06 - 06/15/06	06/27/06 - 06/29/06	07/11/06 - 07/13/06	07/25/06 - 07/27/06	08/09/06 - 08/10/06	08/22/06 - 08/24/06	09/05/06 - 09/07/06	09/19/06 - 09/21/06	10/03/06 - 10/05/06
323	H	H	H	H	H	H	H	H	H	H
340	S/H	S/H	H	B/H	H	H	S/H	H	H	H
405	H	H	H	H	H	H	H	H	H	H
504	H	H	H	H	H	H	H	H	H	H
508	S/H	S/H	H	B/H	H	H	S/H	H	H	H
602	H	H	H	H	H	H	H	H	H	H
609	S/H	S/H	H	B/H	H	H	S/H	H	H	H
704	H	H	H	H	H	H	H	H	H	H
711	S/H	S/H	H	B/H	H	H	S/H	H	H	H
804	H	H	H	H	H	H	H	H	H	H
812	H	H	H	H	H	H	H	H	H	H
902	H	H	H	H	H	H	H	H	H	H
910	S/H	S/H	H	B/H	H	H	S/H	H	H	H
915	S/H	S/H	H	B/H	H	H	S/H	H	H	H
Light 55	H	H	H	H	H	H	H	H	H	H
Vernalis	-	-	-	-	-	-	-	-	-	-
Hood	-	-	-	-	-	-	-	-	-	-
Stockton Port	-	-	-	-	-	-	-	-	-	-

H=Hyalella azteca
 S=Delta smelt
 B=Striped bass
 F=Fathead minnow

Table 2, continued: Sampling Dates and Toxicity Tests Performed

Site	Date									
	10/17/06 - 10/19/06	10/31/06 - 11/02/06	11/14/06 - 11/16/06	11/28/06 - 11/30/06	12/12/06 - 12/13/06	01/30/07- 02/01/07	02/13/07- 2/15/07	2/28/2007- 03/01/07	03/14/07- 03/16/07	03/28/07- 03/29/07
323	H	H	H	H	H	-	-	-	-	-
340	H	H	H	H	H	H	H	H	H	H
405	H	H	H	H	H	H	H	H	H	H
504	H	H	H	H	H	H	H	H	H	H
508	H	H	H	H	H	H	H	H	H	H
602	H	H	H	H	H	H	H	H	H	H
609	H	H	H	H	H	H	H	H	H	H
704	H	H	H	H	H	H	H	H	H	H
711	H	H	H	H	H	H	H	H	H	H
804	H	H	H	H	H	H	H	H	H	H
812	H	H	H	H	H	H	H	H	H	H
902	H	H	H	H	H	H	H	H	H	H
910	H	H	H	H	H	H	H	H	H	H
915	H	H	H	H	H	H	H	H	H	H
Light 55	H	H	H	H	H	H	H	H	H	H
Vernalis	-	-	-	-	-	-	-	-	-	-
Hood	-	-	-	-	-	-	-	-	-	-
Stockton Port	-	-	-	-	-	-	-	-	-	-

H=Hyalella azteca
 S=Delta smelt
 B=Striped bass
 F=Fathead minnow

Table 2, continued: Sampling Dates and Toxicity Tests Performed

Site	Date									
	04/11/07- 04/12/07	04/25/07- 04/27/07	05/08/07- 05/10/07	05/22/07- 05/24/07	06/06/07- 06/08/07	06/20/07- 06/21/07	07/10/07- 07/11/07	07/25/07- 07/26/07	08/08/07- 08/09/07	08/22/07- 08/23/07
323	-	-	-	-	-	-	-	-	-	-
340	H	-	-	-	S/H	S/H	-	S/H	S/H	-
405	H	H	H	H	H	H	H	H	H	H
504	H	H	H	H	H	H	H	H	H	H
508	H	S/H	S/H	S/H	S/H	S/H	H	S/H	S/H	H
602	H	H	H	H	H	H	H	H	H	H
609	H	S/H	S/H	S/H	S/H	S/H	H	S/H	S/H	H
704	H	H	H	H	H	H	H	H	H	H
711	H	S/H	S/H	S/H	S/H	S/H	H	S/H	S/H	H
804	H	H	H	H	H	H	H	H	H	H
812	H	H	H	H	H	H	H	H	H	H
902	H	H	H	H	H	H	H	H	H	H
910	H	H	H	H	H	H	H	H	H	H
915	H	S/H	S/H	S/H	S/H	S/H	H	S/H	S/H	H
Light 55	H	H	H	H	H	H	H	H	H	H
Vernalis	-	S	S	S	S	-	-	S	S	-
Hood	-	S	S	S	S	H	-	S	S	H
Stockton Port	-	-	-	H/F	-	-	-	-	-	-

H=Hyaella azteca
 S=Delta smelt
 B=Striped bass
 F=Fathead minnow

Table 2, continued: Sampling Dates and Toxicity Tests Performed

Site	Date							
	09/04/07 - 09/05/07	09/19/07 - 09/21/07	10/02/07 - 10/04/07	10/16/07 - 10/18/07	10/30/07 - 11/01/07	11/13/07 - 11/15/07	11/27/07 - 11/29/07	12/11/07 - 12/13/07
323	-	-	-	-	-	-	-	-
340	-	-	-	-	-	-	-	-
405	H	H	H	H	H	H	H	H
504	H	H	H	H	H	H	H	H
508	H	H	H	H	H	H	H	H
602	H	H	H	H	H	H	H	H
609	H	H	H	H	H	H	H	H
704	H	H	H	H	H	H	H	H
711	H	H	H	H	H	H	H	H
804	H	H	H	H	H	H	H	H
812	H	H	H	H	H	H	H	H
902	H	H	H	H	H	H	H	H
910	H	H	H	H	H	H	H	H
915	H	H	H	H	H	H	H	H
Light 55	H	H	H	H	H	H	H	H
Vernalis	-	-	-	-	-	-	-	-
Hood	H	H	H	H	H	H	H	H
Stockton Port	-	-	-	-	-	-	-	-

H=Hyalella azteca
 S=Delta smelt
 B=Striped bass
 F=Fathead minnow

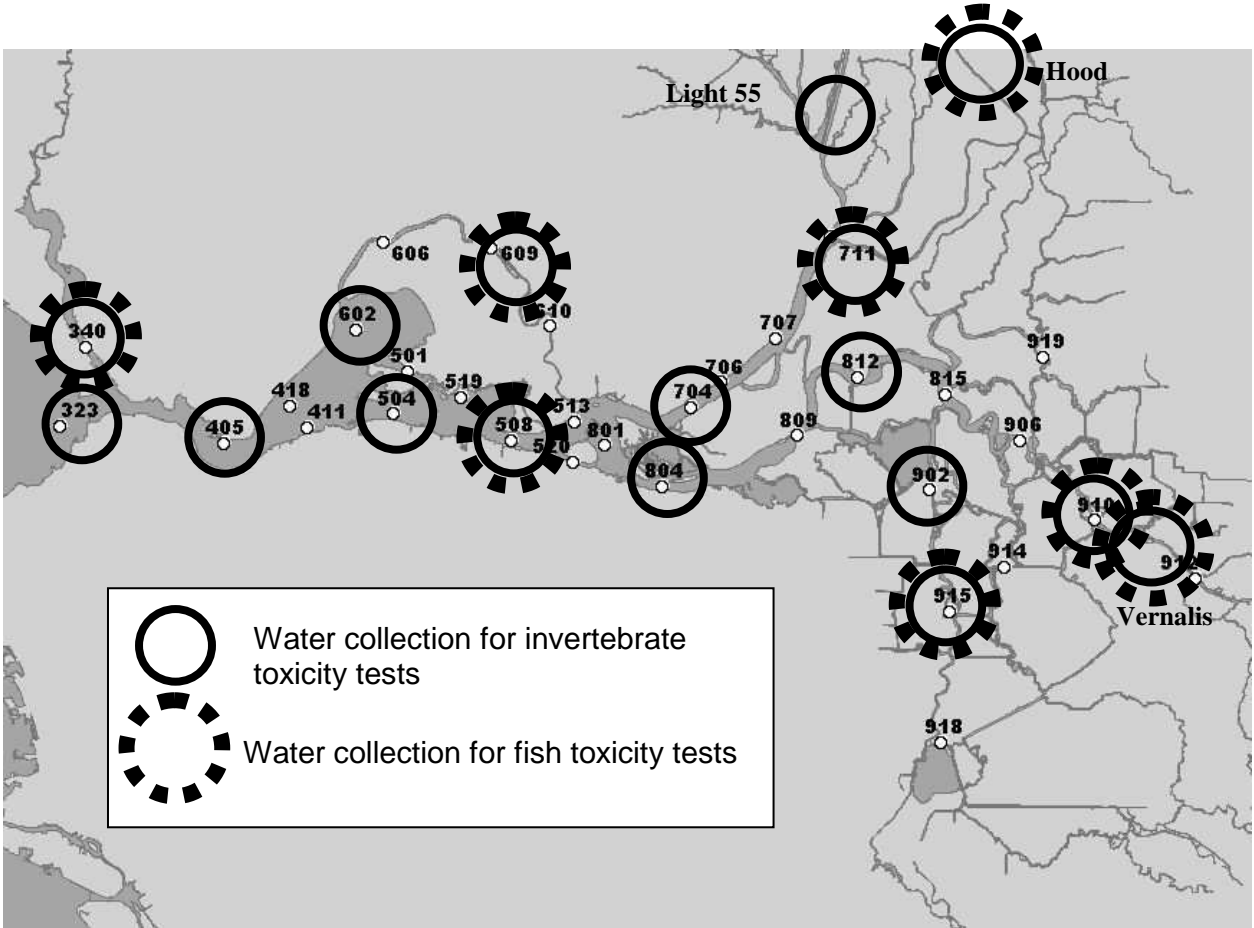


FIGURE 1: Water Toxicity Sampling Locations Based on IEP Summer Townet Survey Stations, 2006-2007 Sampling. Map provided by R. Baxter, CDFG, Bay-Delta Branch.

3.2 Collection of Water Samples

Staff from the UC Davis Aquatic Toxicology Laboratory (UCD ATL) and California Department of Fish and Game (CDFG) collected water samples by boat. Water was pumped into HDPE cubitainers using a standard water pump. Subsurface grab samples were pumped from a depth of approximately 0.5 m into clean, 1-gal amber HDPE cubitainers for invertebrate tests and water chemistry, 1-gal clear HDPE cubitainers for chemical analysis and 5-gal clear HDPE cubitainers for fish tests. Water samples were transported, stored and preserved following protocols outlined in UCD ATL standard operating procedures (SOP), nos. 5-1 and 5-2 (UCD ATL, 2007). All cubitainers for water collection were labeled with the site number, collection date, time and initials of the sampler then rinsed three times with ambient sample water prior to filling. Eight gallons of water were collected from each of the fifteen sites for invertebrate testing, and up to thirty additional gallons were collected for fish testing.

Sediment samples were collected mid-channel using a handheld Stainless Steel Petite Ponar Grab. Sample depth varied from four to twenty-three feet, depending on sample site depth. A 152X152 mm area was sampled for each grab. Approximately 1-liter was collected from the top 2 cm of the sample and placed into clean 500 ml Mason jars. Water for the California Department of Pesticide Regulation (DPR) was collected into certified clean 1-liter narrow mouth amber glass jars with Teflon®-lined lids as subsurface grabs. All samples were placed into an ice chest on wet ice for transport to UCD ATL. Ice was renewed as needed to keep sample temperature at 0-6°C (USEPA 2002). Upon receipt at UCD ATL, water samples were stored in an environmental chamber at $4 \pm 2^\circ\text{C}$. For the single sediment collection event, samples were preserved in a freezer until transfer to the Department of Pesticide Regulation (DPR).

3.3 Water Quality at Sampling Sites

3.3.1 General Water Quality Parameters

Field measurements including pH, specific conductivity (SC), electrical conductivity (EC), dissolved oxygen (DO) and temperature were recorded for each site and sampling time. DO and SC were measured using YSI 85 meters, and pH was measured with a Beckman 240 pH meter. DO/SC and pH meters were calibrated according to the manufacturer's instructions at the start of each field day. Turbidity and ammonia nitrogen were measured within 24 hours of sample receipt at UCD ATL using a Hach 2100P Turbidimeter or a Hach DR/890 Colorimeter with the appropriate Hach AmVer Ammonia Test'N Tube Reagent Set. For ammonia measurements the "low range" test kit (0-2.5 mg/L N) was used first. If the maximum value was exceeded the "high range" test kit (0-50 mg/L N) was used. Unionized ammonia concentrations for all samples were calculated using measured total ammonia-N, as well as field temperature and field pH measurements for each station at the time of sampling. General weather conditions and GPS coordinates were recorded for each site and sampling event. Tables 3 a, b summarize minimum and maximum data by site. Sites are listed in order of increasing maximum EC.

Table 3a. Minimum and maximum water quality parameters measured at sites sampled during 2006 - 2007

Sample	Temperature (°C)		DO (mg/L)		pH		SC (uS/cm)		Turbidity (NTU)	
	Min	Max	Min	Max	Min	Max	Min	Max	Min	Max
323	10.7	21.9	6.7	10.5	7.3	7.8	174	30260	19.8	219.7
340	6.5	25.2	5.8	13.5	7.1	8.6	217	25760	4.9	89.5
405	7.9	23.1	7.0	12.0	6.7	8.2	163	28200	6.1	205.7
504	7.2	24.6	7.0	12.5	6.3	8.1	123	17540	1.4	83.8
508	7.2	24.7	7.3	13.0	6.3	8.1	100	12250	4.2	83.4
602	7.4	22.7	7.5	12.5	6.8	8.1	145	18860	4.8	200.7
609	6.7	26.3	5.1	12.4	6.1	7.9	188	15130	8.6	109.2
704	7.2	25.3	6.8	13.5	6.6	8.2	107	5540	4.6	128.6
711	6.1	25.0	6.8	13.9	6.6	8.3	95	695	2.3	60.8
804	7.2	26.5	6.5	12.9	6.6	8.5	114	5550	4.4	29.0
812	6.7	26.3	6.5	13.6	6.9	8.4	94	832	3.0	13.8
902	5.8	27.2	7.1	12.9	6.3	8.7	132	830	2.2	13.2
910	6.6	28.6	5.3	12.9	6.6	8.3	115	702	3.0	13.0
915	6.6	28.0	6.4	13.3	6.2	8.7	86	721	2.0	10.9
Hood	10.8	23.7	7.0	11.4	7.0	7.6	124	328	2.8	14.1
Light 55	6.4	28.6	6.6	13.9	6.8	8.3	96	534	9.5	68.9
Vernalis	19.6	24.7	9.7	11.0	8.1	8.4	452	587	4.4	13.6
Cache										
Slough@Ulati ¹	20.3	-	8.5	-	7.8	-	272	-	27.7	-
Stockton WWTF ¹	20.3	-	11.0	-	8.8	-	345	-	31.3	-

¹ Site was sampled and tested only once during the project period.

Table 3b. Minimum and maximum measured ammonia, hardness and alkalinity parameters at sites sampled during 2006 - 2007.

Sample	Ammonia Nitrogen (mg/L)		Unionized Ammonia (mg/L)		Hardness (mg/L as CaCO3)		Alkalinity (mg/L as CaCO3)	
	Min	Max	Min	Max	Min	Max	Min	Max
323	0.06	0.20	0.000	0.003	60	3450	62	250
340	0.00	0.33	0.000	0.002	80	3720	57	280
405	0.00	0.49	0.000	0.006	58	3600	49	180
504	0.00	0.26	0.000	0.005	46	1940	30	190
508	0.00	0.24	0.000	0.006	44	1400	46	100
602	0.00	0.27	0.000	0.005	52	3240	48	140
609	0.00	0.27	0.000	0.003	60	1880	52	150
704	0.00	0.30	0.000	0.005	46	618	48	114
711	0.06	0.54	0.000	0.013	44	180	42	82
804	0.00	0.29	0.000	0.008	38	1680	10	88
812	0.00	0.29	0.000	0.005	16	124	36	82
902	0.00	0.24	0.000	0.010	40	272	34	78
910	0.00	0.44	0.000	0.007	38	156	30	104
915	0.00	0.38	0.000	0.006	32	160	34	79
Hood	0.00	0.51	0.000	0.004	52	88	50	86
Light 55	0.00	0.29	0.000	0.012	60	412	60	140
Vernalis Cache	0.00	0.07	0.000	0.006	100	148	58	82
Slough@Ulati ¹	0.20	-	0.005	-	68	-	74	-
Stockton WWTF ¹	0.21	-	0.040	-	80	-	60	-

¹ Site was sampled and tested only once during the project period.

3.3.2 Site-Dependent Differences in Ammonia

Since aquatic organisms in general are sensitive to the toxic effects of ammonia, field data collected during 2006-07 was further analyzed to determine if there were site-dependent differences in ammonia concentrations. Table 4 shows the results of a statistical analysis on data for the entire 2-year period. Tables 5 a-h present results of the data analysis by season.

Overall, total ammonia-N was highest at stations Hood and 711, both on the lower Sacramento River (Table 4). Concentrations at these sites were significantly higher than at most other sampling sites. Other sites with significantly higher ammonia concentrations were 405 (Benicia), 609 (Montezuma Slough), 910 (San Joaquin River), and Light 55 (Sacramento River Deep Water Channel, Table 4). Unionized ammonia concentrations were highest at site 711, Light 55, and site 405.

The season-by-season data analysis presented in Table 5 and Figure 2 shows that site 910 on the San Joaquin River had significantly elevated ammonia levels in 2006 only, whereas sites on the lower Sacramento River (711, Hood, Light 55) had highest concentrations starting in October 2006 until the end of the sampling period in December 2007. In winter (Jan-Mar) 2007, Montezuma Slough (609) and Suisun Bay (602) showed high NH₃ and ammonia-N concentrations, respectively, and site 405 (Benicia) had high concentrations in the summer (Jul-Sep) 2007. The highest mean (+/- standard deviation) quarterly ammonia-N concentrations were recorded at Hood in the spring (Apr-Jun; 0.36+/-0.13 mg/L) and fall (Oct-Dec; 0.36+/-0.05 mg/L) of 2007, and at site 711 in the spring of 2007 (0.29+/-0.12 mg/L). The highest quarterly mean unionized NH₃ concentrations were recorded at Light 55 (0.006+/-0.03 mg/L) and at site 711 in the spring of 2007 (0.007+/-0.004). Other sites with seasonally high NH₃ concentrations were 902 (summer 2006), 602 (winter 2007), and 910 (spring 2007).

Table 4. Ammonia levels in water samples collected at POD sites, 2006 - 2007. Samples indicated by "H" showed significantly higher ammonia levels than some or all of those indicated by "L" (ANOVA with Tukey's multiple comparison procedure, *P* < 0.05). Unionized ammonia data were log transformed prior to analysis to increase homogeneity of variances and reduce outliers.

Site	N	Ammonia Nitrogen (mg/L)			Unionized Ammonia (mg/L)			
		Mean	SD	Significance	N	Mean	SD	Significance
323	14	0.11	0.04	L	14	0.001	0.001	L
340	39	0.08	0.07	L ¹	38	0.001	0.001	L ⁴
405	47	0.13	0.08	H ²	47	0.002	0.001	H ⁴
504	50	0.10	0.06	L	50	0.001	0.001	L ³
508	50	0.10	0.06	L	50	0.001	0.001	L ³
602	49	0.11	0.07	L	49	0.001	0.001	L
609	50	0.12	0.08	H ²	49	0.001	0.001	L ³
704	50	0.11	0.07	L	50	0.001	0.001	L ³
711	50	0.21	0.11	H	49	0.003	0.003	H
804	50	0.09	0.06	L ¹	50	0.001	0.002	L
812	48	0.09	0.06	L ¹	48	0.001	0.001	L ³
902	50	0.06	0.05	L ²	49	0.001	0.002	L ³
910	50	0.15	0.10	H ¹	49	0.002	0.002	L
915	50	0.07	0.07	L ²	49	0.001	0.001	L ⁴
Hood	14	0.28	0.15	H	13	0.002	0.001	-
Light 55	48	0.12	0.08	H ²	47	0.003	0.003	H ³
Vernalis	5	0.03	0.03	L	4	0.002	0.003	-

1. Ammonia nitrogen at 910 was significantly higher than at sites indicated by "L¹" and "L²", and was significantly lower than at sites 711 and Hood.
2. Ammonia nitrogen levels at 405, 609 and Light 55 were significantly higher than at sites 902 and 915, and were significantly lower than at sites 711 and Hood.
3. Unionized ammonia at Light 55 was significantly higher than at sites indicated by "L³", "L⁴" and "L⁵".
4. Unionized ammonia at Hood was only significantly higher than at sites indicated by "L⁴" and "L⁵".
5. Unionized ammonia at 405 was significantly higher than at sites 340 and 915.

Table 5a. Ammonia levels in water samples collected at POD sites, January - March 2006. Samples indicated by "H" showed significantly higher ammonia levels than those indicated by "L" (ANOVA with Tukey's multiple comparison procedure, $P < 0.05$). Unionized ammonia data were log transformed prior to analysis to increase homogeneity of variances and reduce outliers.

Site	N	Ammonia Nitrogen (mg/L)			Unionized Ammonia (mg/L)		
		Mean	SD	Significance	Mean	SD	Significance
323	5	0.10	0.03	L	0.001	0.000	-
340	5	0.13	0.04	L	0.001	0.001	-
405	5	0.10	0.03	L	0.001	0.001	-
504	6	0.10	0.03	L	0.001	0.001	-
508	6	0.09	0.02	L	0.001	0.001	-
602	5	0.11	0.04	L	0.002	0.001	-
609	6	0.16	0.04	L	0.001	0.001	-
704	6	0.09	0.03	L	0.001	0.001	-
711	6	0.11	0.02	L	0.001	0.001	-
804	6	0.09	0.05	L	0.001	0.001	-
812	4	0.08	0.04	L	0.001	0.001	-
902	6	0.07	0.06	L	0.001	0.001	-
910	6	0.29	0.07	H	0.002	0.002	-
915	6	0.11	0.14	L	0.000	0.000	-
Light 55	4	0.08	0.04	L	0.002	0.002	-

Table 5b. Ammonia levels in water samples collected at POD sites, April - June 2006. Samples indicated by "H" showed significantly higher ammonia levels than those indicated by "L" (ANOVA with Tukey's multiple comparison procedure, $P < 0.05$). Unionized ammonia data were log transformed prior to analysis to increase homogeneity of variances and reduce outliers.

Site	N	Ammonia Nitrogen (mg/L)			Unionized Ammonia (mg/L)		
		Mean	SD	Significance	Mean	SD	Significance
323	7	0.13	0.05	-	0.001	0.001	-
340	7	0.11	0.04	-	0.001	0.000	-
405	7	0.12	0.07	-	0.002	0.001	-
504	7	0.07	0.05	-	0.001	0.001	-
508	7	0.08	0.05	-	0.001	0.001	-
602	7	0.07	0.05	-	0.001	0.001	-
609	7	0.11	0.05	-	0.001	0.001	-
704	7	0.06	0.01	-	0.001	0.000	-
711	7	0.15	0.11	H	0.002	0.003	-
804	7	0.05	0.03	L	0.001	0.001	-
812	7	0.07	0.03	-	0.001	0.001	-
902	7	0.05	0.02	L	0.000	0.000	-
910	7	0.13	0.05	-	0.001	0.001	-
915	7	0.07	0.03	-	0.000	0.000	-
Light 55	7	0.05	0.04	L	0.002	0.002	-

Table 5c. Ammonia levels in water samples collected at POD sites, July - September 2006. Samples indicated by "H" showed significantly higher ammonia levels than those indicated by "L" (ANOVA with Tukey's multiple comparison procedure, $P < 0.05$). Unionized ammonia data were log transformed prior to analysis to increase homogeneity of variances and reduce outliers.

Site	N	Ammonia Nitrogen (mg/L)			Unionized Ammonia (mg/L)		
		Mean	SD	Significance	Mean	SD	Significance
323	2	0.08	0.00	-	0.001	0.000	-
340	6	0.04	0.02	-	0.000	0.000	L
405	6	0.10	0.04	-	0.001	0.000	-
504	6	0.09	0.04	-	0.001	0.001	-
508	6	0.08	0.07	L	0.001	0.002	-
602	6	0.08	0.06	-	0.002	0.001	-
609	6	0.06	0.03	L	0.001	0.000	-
704	6	0.10	0.05	-	0.002	0.001	-
711	6	0.13	0.05	-	0.002	0.001	-
804	6	0.07	0.03	L	0.002	0.001	-
812	6	0.06	0.03	L	0.001	0.001	-
902	6	0.03	0.03	-	0.004	0.003	H
910	6	0.20	0.15	H	0.002	0.002	-
915	6	0.05	0.02	L	0.001	0.001	-
Light 55	6	0.04	0.04	L	0.002	0.001	-

Table 5d. Ammonia levels in water samples collected at POD sites, October - December 2006. Samples indicated by "H" showed significantly higher ammonia levels than those indicated by "L" (ANOVA with Tukey's multiple comparison procedure, $P < 0.05$). Unionized ammonia data were log transformed prior to analysis to increase homogeneity of variances and reduce outliers.

Site	N	Ammonia Nitrogen (mg/L)			Unionized Ammonia (mg/L)		
		Mean	SD	Significance	Mean	SD	Significance
340	6	0.04	0.04	L ¹	0.000	0.000	-
405	6	0.19	0.06	-	0.002	0.001	-
504	6	0.13	0.09	L	0.002	0.002	-
508	6	0.14	0.07	L	0.002	0.002	-
602	6	0.17	0.09	-	0.002	0.002	-
609	6	0.18	0.09	-	0.001	0.001	-
704	6	0.16	0.09	-	0.002	0.002	-
711	6	0.32	0.15	H	0.004	0.004	-
804	6	0.17	0.08	-	0.003	0.003	-
812	6	0.16	0.10	-	0.003	0.002	-
902	6	0.11	0.09	L	0.003	0.004	-
910	6	0.18	0.11	-	0.002	0.002	-
915	6	0.11	0.10	L	0.002	0.002	-
Light 55	6	0.24	0.04	H ¹	0.004	0.004	-

¹Ammonia nitrogen at Light 55 was only significantly higher than at site 340.

Table 5e. Ammonia levels in water samples collected at POD sites, January - March 2007. Samples indicated by "H" showed significantly higher ammonia levels than those indicated by "L" (ANOVA with Tukey's multiple comparison procedure, $P < 0.05$). Unionized ammonia data were log transformed prior to analysis to increase homogeneity of variances and reduce outliers.

Site	N	Ammonia Nitrogen (mg/L)			Unionized Ammonia (mg/L)		
		Mean	SD	Significance	Mean	SD	Significance
340	7	0.11	0.12	-	0.001	0.001	L
405	7	0.15	0.05	-	0.001	0.001	-
504	7	0.13	0.07	-	0.001	0.001	-
508	7	0.16	0.05	-	0.001	0.000	-
602	7	0.16	0.06	-	0.002	0.001	H ¹
609	7	0.21	0.06	H	0.001	0.000	-
704	7	0.17	0.09	-	0.001	0.001	-
711	7	0.24	0.10	H	0.002	0.001	H ¹
804	7	0.13	0.06	-	0.001	0.000	-
812	7	0.12	0.06	-	0.001	0.001	-
902	7	0.06	0.04	L	0.000	0.000	L ¹
910	7	0.17	0.06	-	0.001	0.001	-
915	7	0.07	0.04	L	0.000	0.001	L ¹
Light 55	7	0.15	0.07	-	0.002	0.001	L

¹Unionized ammonia levels at 602 and 711 were only significantly greater than at sites 902 and 915.

Table 5f. Ammonia levels in water samples collected at POD sites, April - June 2007. Samples indicated by "H" showed significantly higher ammonia levels than those indicated by "L" (ANOVA with Tukey's multiple comparison procedure, $P < 0.05$). Unionized ammonia data were log transformed prior to analysis to increase homogeneity of variances and reduce outliers.

Site	N	Ammonia Nitrogen (mg/L)			Unionized Ammonia (mg/L)		
		Mean	SD	Significance	Mean	SD	Significance
340	6	0.03	0.05	L ¹	0.000	0.000	L ³
405	6	0.08	0.04	L	0.002	0.001	L
504	6	0.08	0.04	L	0.002	0.001	L
508	6	0.08	0.02	L	0.002	0.001	L
602	6	0.08	0.05	L	0.002	0.001	L
609	6	0.10	0.04	L	0.002	0.001	L
704	6	0.09	0.06	L	0.003	0.002	L
711	6	0.29	0.12	H	0.007	0.004	H
804	6	0.08	0.04	L	0.002	0.001	L
812	6	0.06	0.04	L	0.002	0.002	L
902	6	0.04	0.03	L ¹	0.002	0.002	L
910	6	0.12	0.04	L	0.004	0.002	H ³
915	6	0.04	0.03	L ¹	0.002	0.001	L
Hood	4	0.36	0.13	H	0.003	0.001	H ³
Light 55	6	0.16	0.05	H ^{1,2}	0.006	0.003	H ³
Vernalis	3	0.00	0.01	L ¹	0.000	0.000	L ³

1. Ammonia nitrogen at Light 55 was only significantly greater than at sites indicated by "L¹".
2. Ammonia nitrogen at Light 55 was significantly lower than at sites Hood and 711.
3. Unionized ammonia levels at 910, Hood and Light 55 were only significantly greater than at sites 340 and Vernalis.

Table 5g. Ammonia levels in water samples collected at POD sites, July - September 2007. Samples indicated by "H" showed significantly higher ammonia levels than those indicated by "L" (ANOVA with Tukey's multiple comparison procedure, $P < 0.05$). Unionized ammonia data were log transformed prior to analysis to increase homogeneity of variances and reduce outliers.

Site	N	Ammonia Nitrogen (mg/L)			Unionized Ammonia (mg/L)		
		Mean	SD	Significance	Mean	SD	Significance
340	2	0.06	0.04	L	0.000	0.000	-
405	6	0.09	0.04	H ^{1,2}	0.001	0.001	-
504	6	0.04	0.02	L	0.001	0.001	-
508	6	0.04	0.02	L	0.001	0.001	-
602	6	0.06	0.03	L	0.001	0.001	-
609	6	0.03	0.03	L	0.000	0.001	-
704	6	0.05	0.02	L	0.001	0.001	-
711	6	0.18	0.03	H	0.003	0.003	-
804	6	0.03	0.03	L	0.001	0.002	-
812	6	0.07	0.02	L	0.001	0.002	-
902	6	0.03	0.03	L	0.001	0.002	-
910	6	0.04	0.02	L	0.001	0.001	-
915	6	0.03	0.03	L ¹	0.001	0.002	-
Hood	4	0.10	0.07	L	0.001	0.001	-
Light 55	6	0.05	0.03	L	0.002	0.002	-
Vernalis	2	0.06	0.01	L	-	-	-

1. Ammonia nitrogen at 405 was only significantly greater than at site 915.
2. Ammonia nitrogen at 405 was significantly lower than at site 711.

Table 5h. Ammonia levels in water samples collected at POD sites, October - December 2007. Samples indicated by "H" showed significantly higher ammonia levels than those indicated by "L" (ANOVA with Tukey's multiple comparison procedure, $P < 0.05$). Unionized ammonia data were log transformed prior to analysis to increase homogeneity of variances and reduce outliers.

Site	N	Ammonia Nitrogen (mg/L)			Unionized Ammonia (mg/L)		
		Mean	SD	Significance	Mean	SD	Significance
405	4	0.21	0.19	-	0.003	0.002	H
504	6	0.13	0.06	L	0.001	0.000	-
508	6	0.12	0.05	L	0.001	0.000	-
602	6	0.14	0.04	L	0.001	0.001	-
609	6	0.12	0.10	L	0.000	0.000	-
704	6	0.12	0.09	L	0.001	0.001	-
711	6	0.30	0.05	H ¹	0.002	0.002	H
804	6	0.10	0.07	L	0.001	0.000	-
812	6	0.10	0.03	L	0.001	0.001	-
902	6	0.07	0.05	L	0.000	0.000	L
910	6	0.08	0.02	L	0.001	0.001	-
915	6	0.07	0.05	L	0.001	0.001	-
Hood	6	0.36	0.05	H	0.002	0.001	-
Light 55	6	0.21	0.03	L ¹	0.002	0.002	-

1: Ammonia nitrogen at 711 was not significantly higher than at Light 55.

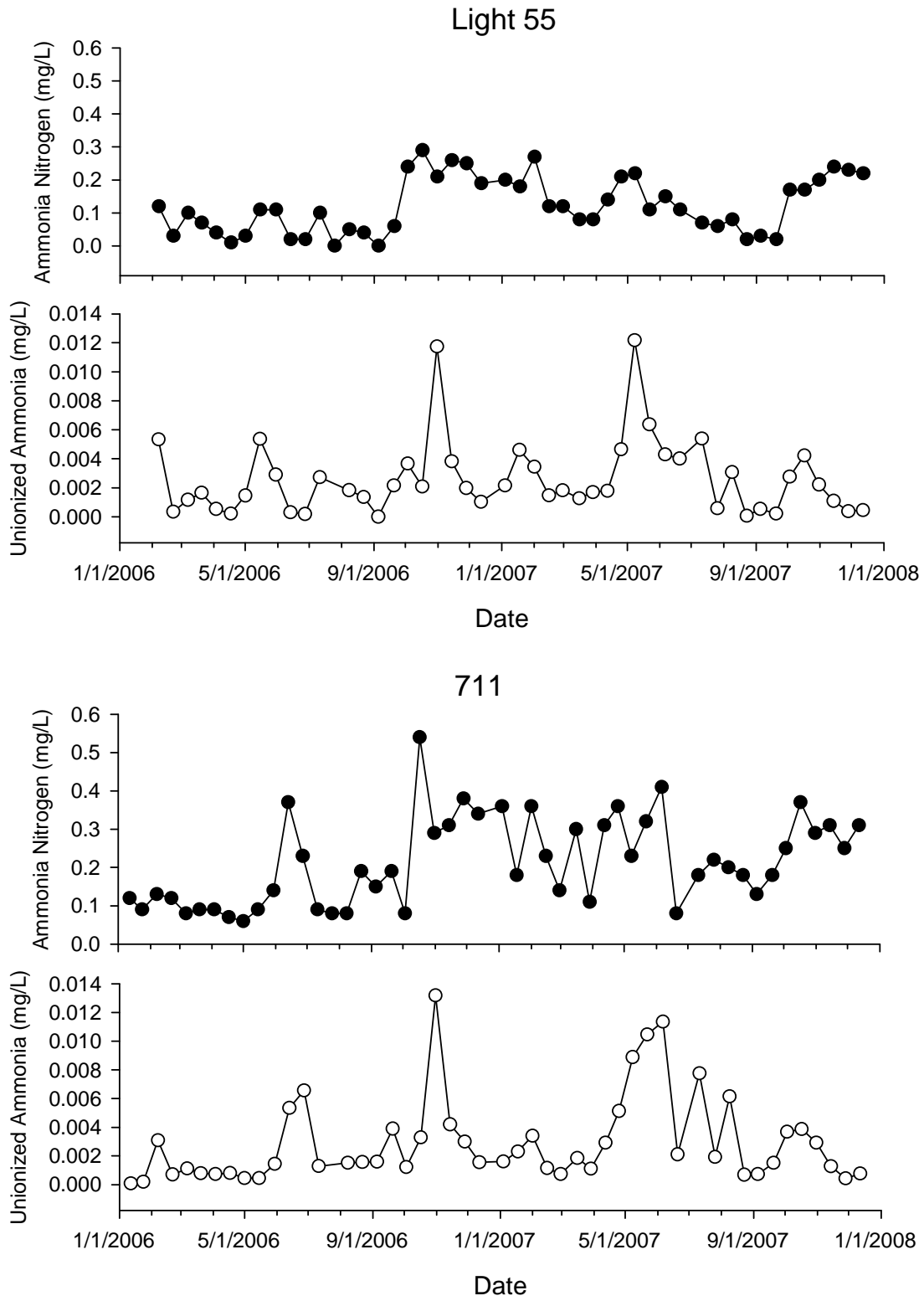


Figure 2 a. Measured ammonia-N and unionized ammonia concentrations during the 2006-2007 project period at Light 55 (Deep Water Ship Channel) and Site 711 (Rio

Vista).

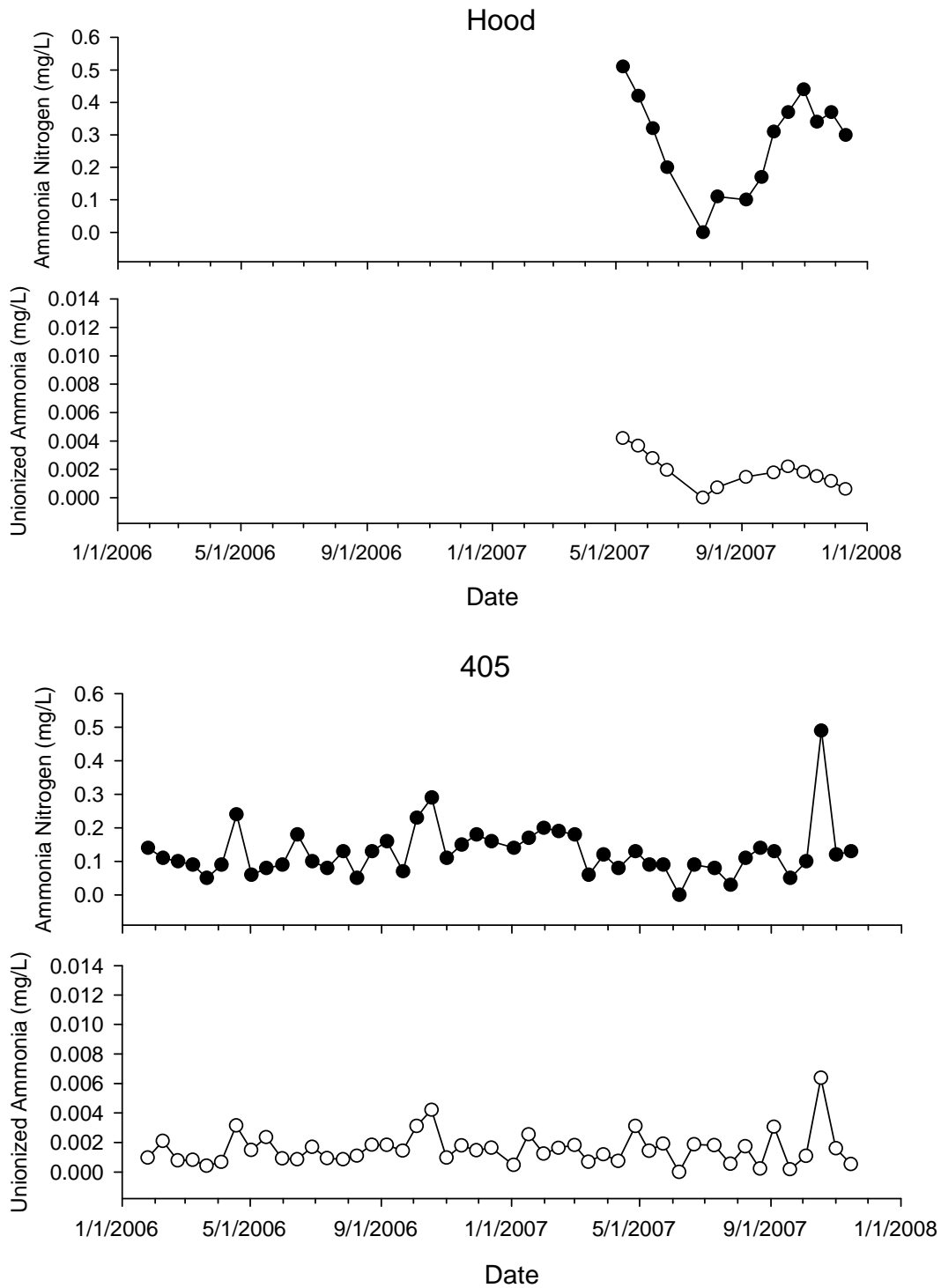


Figure 2 b. Measured ammonia-N and unionized ammonia concentrations during the 2006-2007 project period at Hood (Sacramento River) and Site 405 (Benicia).

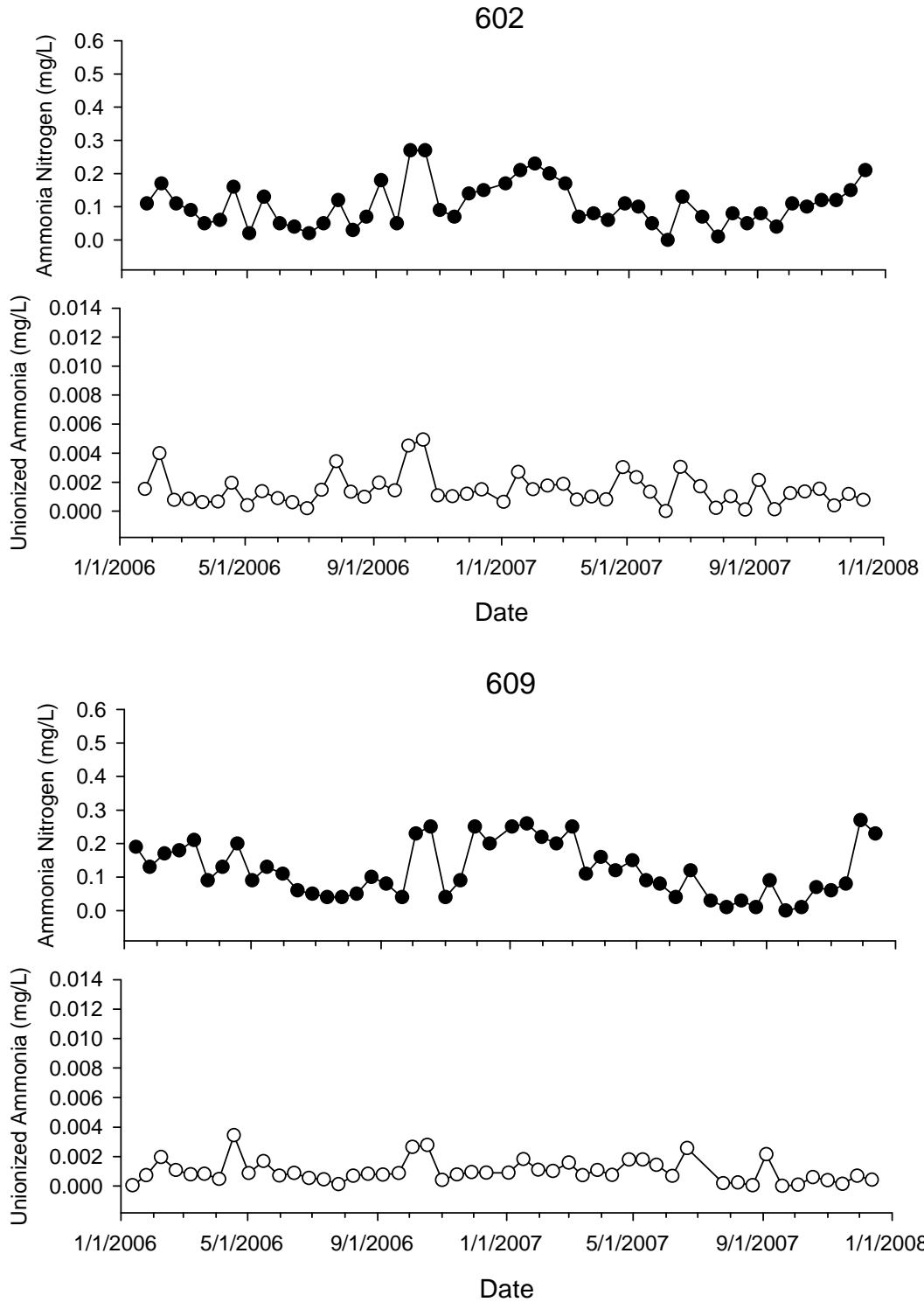


Figure 2 c. Measured ammonia-N and unionized ammonia concentrations during the 2006-2007 project period at sites 602 (Suisun Bay) and Site 609 (Montezuma Slough).

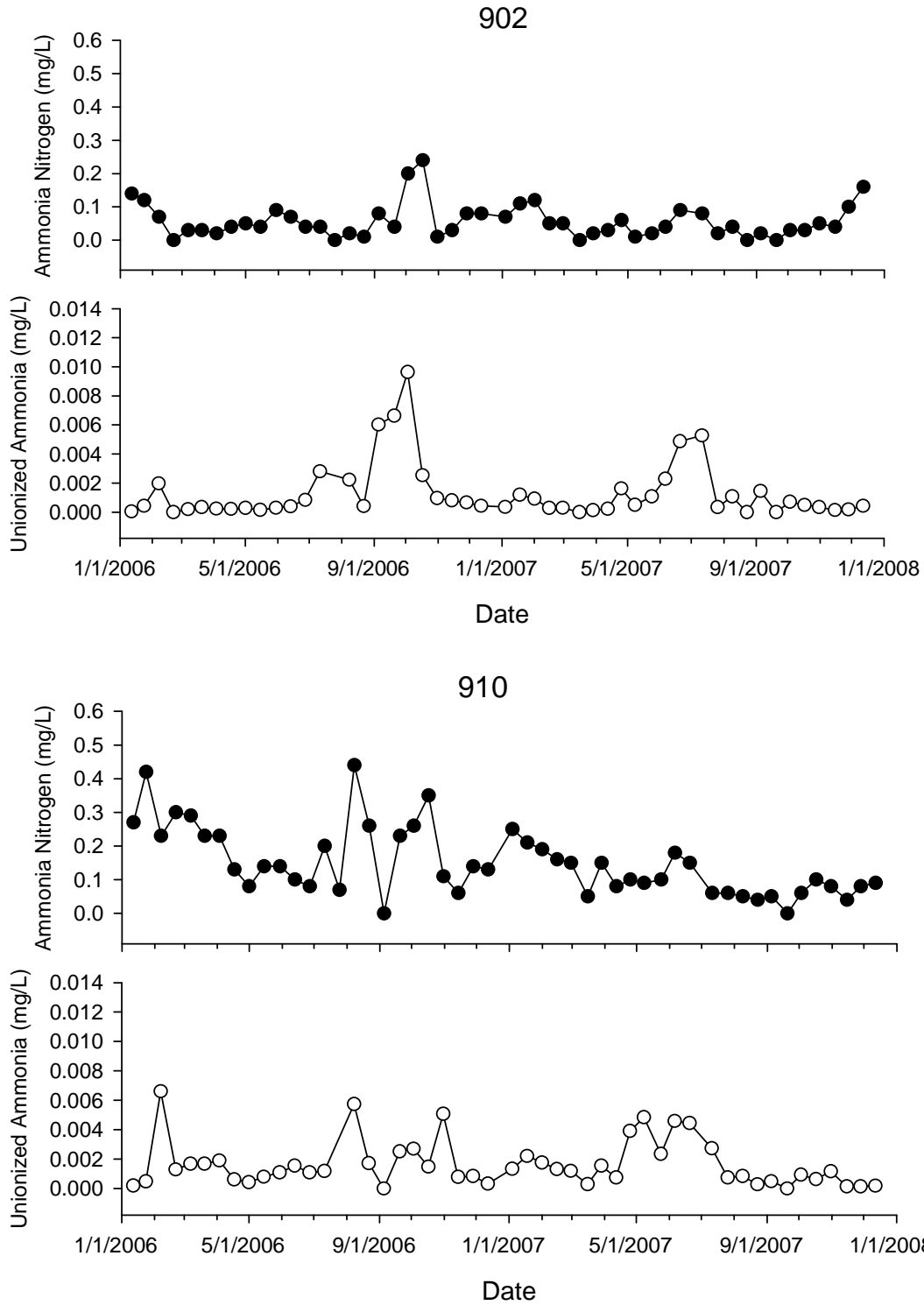


Figure 2 d. Measured ammonia-N and unionized ammonia concentrations during the 2006-2007 project period at sites 902 (Old River) and Site 910 (San Joaquin River).

4. Tests with *Hyalella azteca*

4.1 Field Monitoring

4.1.1 Methods

4.1.1.1 Toxicity Testing

H. azteca were purchased from Aquatic Research Organisms (Hampton, NH). Before initiating bioassays, the water samples were mixed rigorously in the original containers, filtered through a 60- μ m screen, brought to test temperature (23°C) and aerated at a rate of 100 bubbles/min until the dissolved oxygen concentration was approximately 8.5 mg/L. The laboratory control water consists of deionized water amended to US EPA moderately hard standards (DIEPAMHR).

The 10-day tests consisted of four replicate 250 ml glass beakers each containing 100 ml of sample, a one-square-inch piece of nitex screen (a substrate for the *H. azteca* to cling to), and 10 organisms. Tests were initiated with 7 to 14 day old *H. azteca*. Animals in each replicate were fed 1000 μ l of YCT (a mixture of yeast, organic alfalfa and trout chow) on test initiation and days 2, 4, 6, 8, as well as on day 5, when 75% of the test water was renewed. Each series of tests included a standard laboratory control, and if necessary, “high EC controls” and a “low EC control”. “High EC” control water was reconstituted to EPA moderate hardness and the EC adjusted to match the highest EC of the ambient water samples (typically found at site 340, Napa River and 323, San Pablo Bay; and at site 405, Carquinez Strait) with pre-filtered Pacific Ocean seawater obtained from Bodega Bay Marine Laboratory, Bodega Bay, CA. Multiple high EC controls were sometimes conducted in order to have appropriate controls for every sample during sampling events when ambient waters showed a wide range of conductivities. “Low EC” control water was reconstituted to EPA moderate hardness and the EC adjusted to match the lowest EC of the water samples (typically found at site 711, Sacramento River) by diluting with deionized water.

Tests were conducted with and without the addition of piperonyl butoxide (PBO). PBO was added because of its synergistic and antagonistic action with pyrethroid and organophosphate insecticides, respectively. A five parts per million (5 ppm) PBO stock solution was prepared and added to 400 ml of water sample to yield the desired test concentration. Tests were initially conducted with 100 ppb of PBO, which did not affect survival of *H. azteca* (Table 6). However, the concentration was later reduced to 25 ppb because ≥ 50 ppb PBO negatively affected *H. azteca* growth (Table 7). Pairwise analysis of the 2006-07 data revealed no effect of PBO on growth overall, examination of the data by season showed that higher PBO concentrations used in 2006 did affect growth during certain times of the year (Tables 7, 8). The difference in growth was small, and did not affect results of our tests due to relatively high mean standard deviations (MSD) in ambient testing.

Growth in laboratory control water is generally lower than in ambient samples due to the lack of microorganisms naturally present in Delta water. These are obviously an important food source for *H. azteca*. As of 1/04/2007, we added a 1% delta water nutrient concentrate to the laboratory control water (DIEPAMHR) and its counterpart with PBO. The intent of this addition was to more closely match the nutrient and detritus content of control water to that of delta water and to increase the sensitivity of the weight endpoint. As of 2/02/2007, the nutrient concentrate

was also added to the high EC controls and their corresponding PBO treatments. The nutrient concentrate was prepared by centrifuging up to seven Delta water samples with ECs below 1000 mS/cm in a continuous flow centrifuge until 100 times the original concentration was reached. The water used for centrifugation was saved from previous tests after proving to be nontoxic to *H. azteca*. This “nutrient addback” was then added to the control waters and their PBO treatments at 1%, or 1ml to 100 ml of sample waters. An additional control treatment of DIEPAMHR without the “nutrient addback” was included in each test to evaluate the effects of the delta water concentrate on the animals. Our results show that growth of control animals improved considerably when additional natural food was added (Table 8a).

Tests were conducted at a temperature of $23 \pm 2^\circ \text{C}$ with a 16h:8h L:D photoperiod. Mortality was recorded daily, and water was renewed on day 5. On day 10, the surviving *H. azteca* were dried and weighed to determine dry tissue weight per individual and relative growth.

Table 6. Survival of *H. azteca* in a 10-day chronic toxicity test exposed to PBO treated and untreated control waters, some of which were spiked with natural food/organic matter. Differences between treatments with and without PBO were examined by paired t-tests.

Dataset	Control Water	N	Survival (%)		P
			Mean Non-PBO	Mean PBO	
2006 - 2007	No Organic Matter	76	97.2	94.8	0.151
	Organic Matter Added	50	95.5	93.3	0.304

A test to verify if direct toxicity of PBO contributed to the observed effects showed that PBO at a concentration of 25 ppb, used in our tests after 7/27/2006 does not affect 10-day survival or growth of *H. azteca* (Table 7).

Table 7. Summary of 10-day *Hyaella* water column toxicity test initiated on 5/28/07 examining the toxicity of piperonyl butoxide (PBO).

Treatment	Survival (%) ¹		Weight (mg/individual) ¹	
	mean	se	mean	se
DIEPAMHR	90	7.1	0.033	0.003
DIEPAMHR + 5 ppb PBO	90	7.1	0.040	0.006
DIEPAMHR + 10 ppb PBO	100	0.0	0.034	0.002
DIEPAMHR + 15 ppb PBO	100	0.0	0.044	0.005
DIEPAMHR + 20 ppb PBO	100	0.0	0.037	0.003
DIEPAMHR + 25 ppb PBO	98	2.5	0.039	0.005
DIEPAMHR + 50 ppb PBO	98	2.5	0.025	0.004
DIEPAMHR + 100 ppb PBO	98	2.5	0.021	0.001
Weight PMSD = 41.4%				
Weight NOEC = 100 ppb				
Weight EC25 = 42.4 ppb				

¹ Highlighted areas indicate a significant reduction in survival or weight compared to the DIEPAMHR control.

Detailed analysis of our 2006-07 data showed that overall, PBO did not affect *H. azteca* growth (Table 8). When analyzed by season, however, the higher PBO concentrations used during the first part of 2006 (100 ppb until 5/4/2006, and 50 ppb from 5/17-7/14/2006) affected *H. azteca* growth during some seasons. Comparison of final weight data between controls and PBO controls shows that PBO significantly reduced final amphipod weight in tests performed in winter and summer 2006, and in winter 2007. Mean reductions were 28% in winter 2006, when 100 ppb PBO was used, 0% in spring 2006 when 50-100 ppb PBO was used, and 15-20% in summer 2006 and winter 2007, when 25 ppb PBO was used. Tests where PBO addition caused a significant change in final amphipod weight in the control treatments were therefore excluded and samples were not listed as potentially toxic.

Table 8. Final weights of *H. azteca* in a 10-day chronic toxicity test exposed to control water with and without PBO. A control treatment containing natural food/organic matter (“nutrient addback”) was added in 2007. Differences between controls with and without PBO were examined by paired t-tests.

Dataset	Control Water	N	Weight (mg/individual)		P
			Non-PBO Mean	PBO Mean	
2006 - 2007	No Organic Matter	75	0.064	0.060	0.154
	Organic Matter Added	49	0.071	0.068	0.241
Winter 2006	No Organic Matter	10	0.070	0.051	0.003
Spring 2006	No Organic Matter	12	0.074	0.077	0.081
Summer 2006	No Organic Matter	12	0.090	0.072	0.009
Fall 2006	No Organic Matter	12	0.061	0.060	0.728
Winter 2007	Organic Matter Added	14	0.081	0.069	0.047
Spring 2007	No Organic Matter	6	0.049	0.050	0.885
	Organic Matter Added	12	0.072	0.065	0.360
Summer 2007	No Organic Matter	11	0.048	0.051	0.458
	Organic Matter Added	11	0.070	0.072	0.752
Fall 2007	No Organic Matter	12	0.047	0.053	0.096
	Organic Matter Added	12	0.060	0.066	0.150

4.1.1.2 Toxicity Identification Evaluations (TIEs)

TIEs were performed on water samples collected at site 323 on July 12, 2006; site 711 on April 12, 2007; site 405 on August 8, and September 4, 2007; and site Hood on October 2, 2007. Phase I TIEs are generally conducted on samples that cause at least 50% mortality within 7 days to identify the class(es) of contaminant(s) causing the observed toxicity, and involve procedures to either remove or inactivate specific classes of chemicals. After manipulation, the toxicity of a sample is tested and compared to the original water sample. Improved organism performance following TIE manipulation is defined as the absence or a delay of mortality by greater than or equal to 24 hours. Phase I TIEs include manipulations including, but not limited to, air-stripping, low temperature (15°C), Disodium Ethylenediamine Tetraacetate (EDTA) addition, Sodium Thiosulfate (STS) addition, Piperonyl Butoxide (PBO) addition, and solid phase

extraction (C8-SPE).

Heavy metals can be toxic to aquatic species if concentrations exceed threshold levels. EDTA chelates metals, making them unavailable to biota. Three concentrations of EDTA are added to toxic samples and tested along with the appropriate controls. If the toxicant is a metal(s), the unmanipulated sample exhibits high mortality while the sample amended with EDTA causes reduced or no mortality.

PBO decreases toxicity by retarding or preventing formation of the toxicologically active forms of diazinon, chlorpyrifos and other OP insecticides (Bailey *et al.*, 1996). It has no effect on carbofuran, a carbamate insecticide, but potentiates the toxicity of pyrethroid insecticides. PBO is added to the toxic samples for a final concentration of 25 ppb. The unmanipulated sample and the sample amended with PBO are tested along with the appropriate controls in a toxicity test. If the toxicant is a metabolically activated OP insecticide, the unmanipulated test sample will cause high mortality while the test sample amended with PBO results in reduced or no mortality. However, if the toxicant is a carbamate or pyrethroid, both the manipulated and unmanipulated samples will exhibit high mortality.

SPE columns primarily remove non-polar organic chemicals from water samples. A toxic sample is passed through an SPE column and the through-column “rinsate” is tested along with the unmanipulated sample. Control water also is passed through an SPE column and serves as one of the method controls (blank). The adsorbate is then eluted with methanol and the eluate added to control water and tested along with the appropriate method control. If the toxicant is a non-polar organic chemical, the ambient sample and control water amended with methanol eluate will exhibit mortality while the sample passed through the SPE column results in reduced or no mortality.

Air stripping reduces or removes toxicity caused by chemicals such as surfactants, chlorine and/or ammonia from waters. Toxic samples are air stripped and tested along with the appropriate control. If the toxicant is a volatile, the ambient sample exhibits high mortality while the air-stripped sample results in reduced or no mortality. Work performed at UCD ATL documented that air-stripping of a water sample spiked with non-volatile insecticide reduced *C. dubia* mortality.

When ammonia toxicity is suspected based on high ammonia concentrations the pH of the water sample is adjusted to 7.3 and 6.3. At lower pH levels ammonia (NH₃) is converted to ionic ammonium (NH₄⁺), which is less toxic to aquatic organisms, therefore a reduction in toxicity due to lowering of the pH confirms that ammonia was responsible for the observed toxicity.

4.1.1.3 Statistical Analysis

Statistical analysis of *H. azteca* 10-day chronic toxicity data involved three endpoints: 10-day survival, 10-day weight, and 10-day biomass. For each toxicity test a two-part analysis was performed using JMP 5.0.1 (SAS 2003).

First, each unmanipulated (non-PBO) treatment was compared to the non-PBO control. In 2006, we followed modified EPA standard statistical procedures for multiple concentration static renewal toxicity tests (USEPA 2002). Shapiro-Wilk's test and Bartlett's test were used to examine normality of distributions and homogeneity of variances ($\alpha = 0.01$). When non-normal distributions or heteroschedasticity was indicated by these tests, a one-tailed Kruskal-Wallis test was used to determine if significant differences in performance existed among the treatments being compared ($\alpha = 0.05$). When the Kruskal-Wallis test indicated the existence of significant differences, each treatment having a lower mean than the control was compared to the control using Bonferroni-corrected Wilcoxon tests. When normal distributions and homoschedasticity were present, a one-tailed one-way ANOVA was used to determine if significant differences in performance were present ($\alpha = 0.05$). When the ANOVA indicated the existence of significant differences, a Dunnett's multiple comparison procedure was performed to determine which treatments showed significant differences from the control (one-tailed $\alpha = 0.05$). In tests containing high or low conductivity samples (high EC > 10,000 uS/cm; low EC < 100 uS/cm) and a high or low conductivity control treatment, statistics were performed separately for the normal conductivity subset of samples and the high or low conductivity subset.

Second, each sample and control water treatment was compared to its PBO treated counterpart by a full factorial two-way ANOVA (two-tailed $\alpha = 0.05$). The three terms in the ANOVA were 1) the identity of test water, 2) the presence or absence of PBO and 3) an interaction term between test water and PBO presence. When there was a significant overall effect of PBO or interaction effect, a Tukey's multiple comparison procedure was performed to identify if a significant difference existed between any control or test water and its PBO treated counterpart, and to identify if any PBO-treated sample showed a significant decrease in survival or weight relative to the PBO-treated control of the most appropriate conductivity.

In 2007, we changed statistical methods to maximize and standardize test sensitivity and to allow the calculation of meaningful minimum significant differences (MSDs) for all tests. Instead of using a modification of USEPA statistics intended for multiple concentration tests, we used one-way ANOVA and Tukey's multiple comparison procedure to evaluate all comparisons among waters not treated with PBO. Tukey's multiple comparison procedure has greater statistical sensitivity than most of the methods involved in the USEPA protocol, and it has the advantage of evaluating of all possible pairwise comparisons between treatments, instead of being limited to comparing each treatment to one control. The USEPA protocol requires that data are tested for normality and homogeneity of variance before being tested using ANOVA. However, Zar (1996) reports that tests for homogeneity of variance perform poorly and are not recommended for testing the underlying assumptions of ANOVA, and reports that ANOVA is reliable for multisample testing among means even in cases of substantial heterogeneity of variances or considerable deviations from normality. Therefore, data were not tested for normality or homogeneity of variance before being tested with ANOVA and Tukey's procedure. Significant reductions in survival and weight in unmanipulated (ambient) samples were evaluated relative to the control with the most appropriate conductivity. The statistical evaluation of PBO-treated water samples did not change in 2007; we continued to use the two-way ANOVA protocol outlined above. We calculated MSDs for all one-way and two-way ANOVA

Tukey's tests to track the sensitivity of the endpoints over the course of the year.

Methods used in the analysis of long-term patterns and trends included pairwise correlations, ANOVA, ANCOVA, MANOVA, linear regression, and polynomial regression models performed in JMP 5.0.1. Polynomial regression showed that conductivity affected both the survival and weight of *H. azteca* in 10-day chronic toxicity tests. Therefore, conductivity was included in models constructed to examine the effects of factors such as site, season and PBO treatment on *H. azteca* survival and weight.

Many samples and controls were simultaneously tested with and without the addition of PBO. This enabled us to consider PBO as a within-subjects (or repeated measures) factor in models designed to examine the effects of PBO. Paired t-tests were used to examine the effects of PBO in normal EC control water. MANOVA models with non-PBO and PBO-treated toxicity endpoints as paired response variables were used to examine the effects of PBO in ambient water samples and in high conductivity controls while controlling for the effects of differences in conductivity.

PBO Effects on Weight: PBO was shown to cause significant decreases in *H. azteca* weight when added to some ambient delta water samples in 2006, but also caused small but frequent decreases in *H. azteca* weight in control waters. A large number of delta water samples were tested during this study, and the question arises if the significant decreases in *H. azteca* weight due to PBO addition could have been found randomly due to the same effects seen in the controls, with the greater number of significant effects occurring due to the greater sample size of ambient waters tested. To address this possibility, changes in *H. azteca* weight in control waters with PBO addition were examined separately in each season. The mean and standard deviation of weight change in the control waters for each season were used to calculate a z-score for each ambient sample showing a significant reduction in weight with PBO addition. This z-score allowed the calculation of the probability and numbers of samples expected to show the level of weight reduction showed by the ambient sample, if the effects of PBO in ambient samples conform to the null model of the effects of PBO in control waters. The numbers of samples expected by the null model to show given levels of reduction in weight were compared to the numbers of samples actually observed at those levels of weight reduction to reveal if the ambient samples showed a greater extent of weight reduction with PBO addition than would be expected due solely to the pattern of weight reduction seen in the controls.

4.1.1.4 Analytical Chemistry

Water samples for analytical chemistry were collected at each sampling site and sampling event using acid-cleaned, amber water bottles, transported on ice and stored at 4°C. If a sample noticeably affected survival or growth of *H. azteca*, samples were submitted to the California Department of Fish and Game – Water Pollution Laboratory, Rancho Cordova, CA, for chemical analysis. As of June 20/21, 2007, 10 mL dichloromethylene (DCM) were added to one 1-L sample upon receipt at UCD ATL to prevent possible degradation of pyrethroid insecticides during storage.

4.1.2 Results

A total of 693 water samples were collected for toxicity testing with *H. azteca* during the project period January 1, 2006 to December 31, 2007. Results of the toxicity tests are summarized in Tables 9 a-c and 10 a, b below. Detailed results and water chemistry data are shown in Appendix A, Tables A2-A107.

4.1.2.1 Acute Toxicity to *H. azteca* - Effects on 10-d Survival

During the 2006-2007 period, a total of fifteen water samples (2.2% of total samples tested) were acutely toxic causing a significant reduction in amphipod survival (Table 9 a). Table 10 a shows from which sites these samples were collected, and the relative site-specific percentage of samples showing toxic effects, since sample numbers varied for some sites. A more detailed listing of results including the dates when samples were collected is presented in Table 11 a. Most of the acutely toxic samples were from sites in the lower Sacramento River (Hood, 711), the Deep Water Ship Channel (Light 55) and site 405 (Benicia). In addition, one sample collected on 7/10/07 from site 602 (Suisun Bay) and one sample from site 323 (7/12/06, San Pablo Bay) were acutely toxic. The majority of toxic samples (93.3%) were collected in 2007 (Table 9 b), mostly during the second half of the year (July-December; Table 9 c). Of all samples tested in 2006, only 0.3% exhibited acute toxicity, while 4.1% of samples tested in 2007 were toxic. Since 2006 was a year with high precipitation and river flows, and flows are generally higher in the first part of the year, this pattern suggests an inverse relationship of toxicity with flows.

PBO Effect on 10-d Survival: Significant changes in acute toxicity due to PBO addition to the ambient samples were seen in seven additional samples (1% of total samples tested) from the Sacramento River at Hood, sites 711, 704, Light 55, 340 (Napa River), 405 (Benicia) and 323 (San Pablo Bay). Toxicity was reduced due to PBO addition indicating the possible presence of organophosphate insecticides in samples collected from Hood on Oct 2, and Oct 30, 2007, and from Light 55 on Oct 31, 2007. Two samples collected on Apr 12, 2007 from site 711, and on Feb 1, 2007 from Light 55 showed a trend towards improved survival due to PBO addition. PBO increased toxicity, indicating the possible presence of pyrethroid insecticides, in samples collected on Jan 25, 2006 at site 323, Aug 22, 2006 at site 711, Mar 29, 2007, at site 340, and Aug 8, 2007 at site 405. In addition, PBO addition significantly reduced 48-hour survival in samples collected on July 10, 2007 from sites 804, 504 and 508 (Table A81-1), three adjacent field sites, suggesting that PBO-synergized chemicals such as pyrethroid insecticides may have been present. Only 0.047 ug/L piperonyl butoxide, a synergist used in pyrethroid pesticide formulations, was detected at site 804. Toxicity seen in samples collected on February 28, 2007 at site 711, Jul 25, 2007 at site 704, Oct 16 and Nov 13, 2007 at Hood, and Nov 28, 2007 at Light 55 remained unchanged after addition of PBO.

4.1.2.2 Chronic Toxicity to *H. azteca* - Effects on 10-d Growth

Only one sample (site 323) reduced *H. azteca* growth (Tables 9 a, 10 b). In general, this

endpoint was not a sensitive indicator of toxicity due to the variable size of the organisms, and the variability in food content between Delta water samples from different sites. Table 10 b shows from which site this sample was collected. A more detailed listing of results including the dates when samples were collected is presented in Table 11 b.

PBO Effect on 10-d Growth: Addition of PBO to the ambient sample resulted in a significant reduction or increase in amphipod growth (relative weight at test termination) when compared to the ambient sample in a total of 33 water samples (4.9% of samples tested; Table 9 a), independent of control growth. PBO addition led to increased growth in 3, and decreased growth in 29 samples. A significant reduction in growth compared to the ambient sample suggests the presence of pyrethroid insecticides at chronically toxic concentrations. A significant increase in growth suggests the presence of organophosphate insecticides. While significant PBO effects on amphipod growth were detected in 14 samples in 2006 (4.1%), a total of 19 (5.7%) showed this effect in 2007 (Table 9 b, c).

Water samples where PBO addition resulted in a reduction in growth were primarily collected from sites in the South-Eastern Delta (902, 910, 915), the lower Sacramento River (Light 55, 711) and Suisun Bay (609, 602, 508). Patterns where several neighboring sites sampled on the same date triggered the same response in bioassay organisms were seen repeatedly (Table 11 b). Most samples where a PBO effect on amphipod growth was detected were collected in the spring or summer period. Three water samples where PBO addition resulted in an increase in growth were collected from sites 902, 910 and 812 on June 6, 2007.

Table 9 a. Total numbers of samples tested using the 10-day *H. azteca* water column test, and samples showing toxicity, January 1, 2006– December 31, 2007¹. Numbers of statistical comparisons of controls to controls containing PBO are given for reference.

Sample Type	Comparison	Number of Samples	Number of Samples Affecting Survival		Number of Samples Affecting Weight	
			Reduced	Increased	Reduced	Increased
Ambient	v. EC-specific Control	693	15	NA	1	NA
Ambient with PBO	v. EC-specific PBO Control	673	8	NA	4	NA
Ambient with PBO	v. Ambient	677	4	3	30	3
PBO Control	v. Non-PBO Control	125	2	1	2	2
High EC PBO Control	v. High EC Non-PBO Control	84	4	1	1	0

¹ Quality Assurance samples are not included

Table 9 b. Total number of samples tested using the 10-day *H. azteca* water column test, and number of samples showing toxicity by year, 2006 and 2007¹.

Year	Sample Type	Comparison	Number of Samples	Number of Samples Affecting Survival		Number of Samples Affecting Weight	
				Reduced	Increased	Reduced	Increased
2006	Ambient	v. EC-specific Control	353	1	NA	1	NA
	Ambient with PBO	v. EC-specific PBO Control	338	1	NA	1	NA
	Ambient with PBO	v. Ambient	342	2	0	14	0
2007	Ambient	v. EC-specific Control	340	14	NA	0	NA
	Ambient with PBO	v. EC-specific PBO Control	335	7	NA	3	NA
	Ambient with PBO	v. Ambient	335	2	3	16	3

¹ Quality Assurance samples are not included

Table 9 c. Total number of samples tested using the 10-day *H. azteca* water column test, and number of samples showing the number of toxic samples during 2006-2007 listed by half year¹.

Time Period	Sample Type	Comparison	Number of Samples	Number of Samples Affecting Survival		Number of Samples Affecting Weight	
				Reduced	Increased	Reduced	Increased
Jan - Jun 2006	Ambient	v. EC-specific Control	187	0	NA	1	NA
	Ambient with PBO	v. EC-specific PBO Control	172	1	NA	1	NA
	Ambient with PBO	v. Ambient	172	1	0	3	0
Jul - Dec 2006	Ambient	v. EC-specific Control	166	1	NA	0	NA
	Ambient with PBO	v. EC-specific PBO Control	166	0	NA	0	NA
	Ambient with PBO	v. Ambient	170	1	0	11	0
Jan - Jun 2007	Ambient	v. EC-specific Control	184	3	NA	0	NA
	Ambient with PBO	v. EC-specific PBO Control	179	2	NA	3	NA
	Ambient with PBO	v. Ambient	179	0	0	13	3
Jul - Dec 2007	Ambient	v. EC-specific Control	156	11	NA	0	NA
	Ambient with PBO	v. EC-specific PBO Control	156	5	NA	0	NA
	Ambient with PBO	v. Ambient	156	2	3	3	0

¹ Quality Assurance samples are not included

Table 10 a. Minimum and maximum *H. azteca* survival in 10-day chronic water column toxicity tests performed during 2006 – 2007, and site-specific percentage of toxic samples.

Sampling Site	Ambient Samples: Survival (%)		Number and % Toxic Samples	Survival (%) After PBO Addition		PBO-Effect		
	Min	Max		Min	Max	# Signif. Reduction vs Control	# Signif. Increase in Toxicity	# Signif. Decrease in Toxicity
Hood	43.0	97.5	3 (38%)	67.5	100.0	2 (25%)	0	2* (25%)
POD 711	63.1	100.0	3 (%)	43.3	100.0	1 (2%)	1(2%)	0
POD 910	87.5	100.0	0	66.7	100.0	0	0	0
POD 915	78.0	100.0	0	60.6	100.0	0	0	0
POD 902	66.0	100.0	0	69.5	100.0	0	0	0
POD 812	62.4	100.0	0	43.3	100.0	0	0	0
Light 55	76.9	100.0	3 (6%)	58.8	100.0	1 (2%)	0	1 (2%)
POD 704	84.5	100.0	0	15.6	100.0	1 (2%)	0	0
POD 804	87.5	100.0	0	0.0	100.0	0	0	0
POD 508	80.2	100.0	0	0.0	100.0	0	0	0
POD 609	70.0	100.0	0	70.0	100.0	0	0	0
POD 504	75.6	100.0	0	0.0	100.0	0	0	0
POD 602	25.4	100.0	1 (2%)	5.0	100.0	0	0	0
POD 340	46.0	100.0	0	30.7	100.0	0	1 (3%)	0
POD 405	0.0	100.0	4 (9%)	0.0	100.0	0	1 (2%)	0
POD 323	4.5	100.0	1 (7%)	14.8	100.0	1 (7%)	1 (7%)	0

* The difference between PBO-treated and ambient water sample was not significant. Both samples showed reduced survival without PBO addition, but in one case the reduction vs. control was not statistically significant.

Table 10 b. Minimum and maximum *H. azteca* final weights after 10-day chronic water column toxicity tests performed during 2006 – 2007, and site-specific percentage of affected samples.

Sample	Non-PBO Weight (mg dry wt./individual)			Weight (mg dry wt./individual) After PBO Addition				
	Min	Max	# Signif. Reduction vs Control	Min	Max	# Signif. Reduction vs Control	# Signif. Increase in Toxicity	# Signif. Decrease in Toxicity
Hood	0.045	0.093	0	0.035	0.066	0	0	0
POD 711	0.043	0.159	0	0.031	0.144	0	3 (6%)	0
POD 910	0.036	0.199	0	0.047	0.168	0	3 (6%)	1 (2%)
POD 915	0.049	0.162	0	0.050	0.151	0	3 (6%)	0
POD 902	0.031	0.182	0	0.048	0.143	0	4 (8%)	1 (2%)
POD 812	0.033	0.187	0	0.033	0.173	1 (2%)	1 (2%)	1 (2%)
Light 55	0.042	0.182	0	0.040	0.158	0	2 (4%)	0
POD 704	0.038	0.192	0	0.045	0.178	0	0	0
POD 804	0.041	0.177	0	0.047	0.172	0	0	0
POD 508	0.032	0.156	0	0.037	0.146	0	3 (6%)	0
POD 609	0.039	0.182	0	0.030	0.204	0	3 (6%)	0
POD 504	0.021	0.182	0	0.032	0.162	0	3 (6%)	0
POD 602	0.026	0.164	0	0.024	0.166	0	2 (4%)	0
POD 340	0.020	0.195	0	0.007	0.180	0	1 (3%)	0
POD 405	0.017	0.153	0	0.023	0.179	0	1	0
POD 323	0.047	0.168	1 (7%)	0.003	0.122	0	1 (7%)	0

Table 11 a. *H. azteca* Survival: Water samples that significantly reduced *H. azteca* survival in 10-day water column toxicity tests performed January 1, 2006 – December 31, 2007.

Sample	Collection Date	Test Date	EC Specific Control			Ambient Sample				
			Mean Non-PBO Survival (%)	Mean PBO Survival (%)	Signif. PBO Effect ²	Mean Non-PBO Survival (%) ¹	Mean PBO Survival (%) ¹	Signif. PBO Effect ²		
POD 323	1/25/2006	1/26/2006	96	97	No	94	51	↓Yes		
POD 323	7/12/2006	7/13/2006	88	-	-	34*	41	No		
POD 711	8/22/2006	8/24/2006	100	97	No	98	43	↓Yes		
Light 55	2/1/2007	2/2/2007	100	83	No	77	95	No		
POD 711	2/28/2007	3/1/2007	100	95	No	78	76	No		
POD 704	3/29/2007	3/30/2007	98	100	No	93	84	No		
POD 711	4/12/2007	4/13/2007	100	84	No	63*	87	No		
POD 602	7/10/2007	7/12/2007	93	-	-	49	-	-		
POD 405	7/10/2007	7/12/2007	58	-	-	3	-	-		
POD 340	7/25/2007	7/27/2007	83	73	No	67	44	↓Yes		
POD 405	8/8/2007	8/9/2007	31	5	↓Yes	56*	29	↓Yes		
POD 405	8/22/2007	8/23/2007	75	48	No	30	28	No		
POD 711	8/23/2007	8/24/2007	100	100	No	88	98	No		
POD 405	9/4/2007	9/5/2007	38	5	No	13*	15	No		
Hood	10/2/2007	10/4/2007	97	98	No	43*	89	↑Yes		
POD 405	10/4/2007	10/5/2007	98	97	No	76	77	No		
Hood	10/16/2007	10/18/2007	97	100	No	86	84	No		
Hood	10/30/2007	11/1/2007	100	98	No	82	91	↑Yes		
Light 55	10/31/2007	11/1/2007	100	98	No	90	100	↑Yes		
Hood	11/13/2007	11/15/2007	98	97	No	76	68	No		
Light 55	11/28/2007	11/29/2007	97	98	No	82	75	No		

¹ Highlighted cells indicate ambient sample treatments showing significantly lower survival than the EC specific control.

² Highlighted cells indicate significant differences between the unmanipulated and PBO-treated water samples; arrows indicate ↓a reduction in survival, and ↑an increase in survival due to PBO.

* TIEs were performed on these samples

Table 11 b. *H. azteca* Growth: Water samples that significantly reduced or enhanced *H. azteca* growth during 10-day water column toxicity tests performed January 1, 2006 – December 31, 2007.

Sample	Collection Date	Test Date	EC Specific Control			Ambient Sample		
			Mean Non-PBO Weight (mg/individual)	Mean PBO Weight (mg/individual)/ (% non-PBO)	Signif. PBO Effect ²	Mean Non-PBO Weight (mg/individual) ¹	Mean PBO Weight (mg/individual) ¹ / (% non-PBO)	Signif. PBO Effect ²
POD 504	3/21/2006	3/22/2006	0.076	0.056 (74%)	No	0.122	0.078 (64%)	↓Yes
POD 915	4/17/2006	4/18/2006	0.085	0.056 (66%)	No	0.162	0.077 (48%)	↓Yes
POD 323	6/14/2006	6/15/2006	0.083	-	-	0.047	0.063	No
POD 812	6/29/2006	6/30/2006	0.054	0.167 (310%)	Yes	0.187	0.033 (17.7%)	↓Yes
POD 323	7/12/2006	7/13/2006	0.028	-	-	0.132	0.037 (28%)	↓Yes
POD 609	8/23/2006	8/24/2006	0.090	0.076 (84%)	No	0.106	0.048 (45.3%)	↓Yes
POD 711	8/22/2006	8/24/2006	0.090	0.076 (84%)	No	0.105	0.039 (37.1%)	↓Yes
POD 902	8/23/2006	8/24/2006	0.090	0.076 (84%)	No	0.124	0.059 (47.6%)	↓Yes
Light 55	8/22/2006	8/24/2006	0.090	0.076 (84%)	No	0.138	0.065 (47.1%)	↓Yes
POD 405	9/21/2006	9/22/2006	0.064	0.045 (70%)	No	0.101	0.054 (53.5%)	↓Yes
POD 504	9/21/2006	9/22/2006	0.064	0.045 (70%)	No	0.115	0.054 (47%)	↓Yes
POD 508	9/21/2006	9/22/2006	0.064	0.045 (70%)	No	0.119	0.065 (54.6%)	↓Yes
POD 711	10/3/2006	10/5/2006	0.072	0.065 (90.3%)	No	0.069	0.041 (59.4%)	↓Yes
POD 902	10/3/2006	10/5/2006	0.072	0.065 (90.3%)	No	0.103	0.072 (69.9%)	↓Yes
POD 910	10/3/2006	10/5/2006	0.072	0.065 (90.3%)	No	0.109	0.078 (71.6%)	↓Yes
POD 405 ³	6/28/2006	6/30/2006	0.054	0.167	Yes	0.064	0.179	↑Yes

¹ Highlighted cells indicate ambient sample treatments showing significantly lower survival than the EC specific control.

² Highlighted cells indicate significant differences between the unmanipulated and PBO-treated water samples; arrows indicate ↓a reduction in growth, and ↑an increase in growth due to PBO.

³ Growth effects of PBO were seen in controls, thus effect in ambient sample was not attributed to toxic contaminants.

Table 11b (continued).

Sample	Collection Date	Test Date	EC Specific Control			Ambient Sample		
			Mean Non-PBO Weight (mg/individual)	Mean PBO Weight (mg/individual) / (% non-PBO)	Signif. PBO Effect ²	Mean Non-PBO Weight (mg/individual) ¹	Mean PBO Weight (mg/individual) ¹ / (% non-PBO)	Signif. PBO Effect ²
POD 910	1/4/2007	1/5/2007	0.085	0.057 (67.1%)	No	0.111	0.063 (56.8%)	↓Yes
POD 504	1/17/2007	1/18/2007	0.076	0.045 (59.2%)	No	0.088	0.040 (45.5%)	↓Yes
POD 910	2/1/2007	2/2/2007	0.107	0.090 (84.1%)	No	0.137	0.097 (70.8%)	↓Yes
POD 915	2/1/2007	2/2/2007	0.107	0.090 (84.1%)	No	0.137	0.092 (67.2%)	↓Yes
POD 340	2/13/2007	2/15/2007	0.053	0.052 (98.1%)	No	0.098	0.064 (65.3%)	↓Yes
Light 55	2/15/2007	2/16/2007	0.049	0.063 (128.6%)	No	0.097	0.040 (41.2%)	↓Yes
POD 902	2/15/2007	2/16/2007	0.049	0.063 (128.6%)	No	0.103	0.052 (50.5%)	↓Yes
POD 915	2/28/2007	3/1/2007	0.090	0.078 (86.7%)	No	0.116	0.065 (56.0%)	↓Yes
POD 508	3/1/2007	3/2/2007	0.084	0.059 (70.2%)	No	0.101	0.061 (60.4%)	↓Yes
POD 602	3/14/2007	3/14/2007	0.091	0.114 (125.3%)	No	0.142	0.106 (74.1%)	↓Yes
POD 609	3/14/2007	3/14/2007	0.091	0.114 (125.3%)	No	0.149	0.114 (76.5%)	↓Yes
POD 609	4/11/2007	4/18/2007	0.106	0.081 (76.4%)	No	0.125	0.084 (68.0%)	↓Yes
POD 508	5/23/2007	5/24/2007	0.067	0.042 (62.7%)	No	0.090	0.040 (44.4%)	↓Yes
POD 812	6/6/2007	6/7/2007	0.051	0.048 (94.1%)	No	0.060	0.110 (183.3%)	↑Yes
POD 902	6/6/2007	6/7/2007	0.051	0.048 (94.1%)	No	0.045	0.115 (255.6%)	↑Yes
POD 910	6/6/2007	6/7/2007	0.051	0.048 (94.1%)	No	0.061	0.104 (170.5%)	↑Yes
POD 602	9/19/2007	9/20/2007	0.055	0.038 (69.1%)	No	0.054	0.024 (44.4%)	↓Yes
POD 902	10/17/2007	10/18/2007	0.056	0.051 (91.1%)	No	0.095	0.060 (63.2%)	↓Yes
POD 711	10/31/2007	11/1/2007	0.057	0.082 (143.9%)	No	0.084	0.042 (50.0%)	↓Yes
POD 704 ³	1/18/2007	1/19/2007	0.071	0.028 (39.4%)	Yes	0.102	0.054 (52.9%)	↓Yes
POD 711 ³	6/20/2007	6/21/2007	0.115	0.157 (136.5%)	Yes	0.088	0.144 (163.6%)	↑Yes
POD 812 ³	6/20/2007	6/21/2007	0.115	0.157 (136.5%)	Yes	0.121	0.101 (83.5%)	No
POD 910 ³	6/20/2007	6/21/2007	0.115	0.157 (136.5%)	Yes	0.138	0.102 (73.9%)	No
POD 915 ³	6/20/2007	6/21/2007	0.115	0.157 (136.5%)	Yes	0.136	0.097 (71.3%)	No

¹ Highlighted cells indicate ambient sample treatments showing significantly lower survival than the EC specific control.

² Highlighted cells indicate significant differences between the unmanipulated and PBO-treated water samples; arrows indicate ↓a reduction in growth, and ↑an increase in growth due to PBO.

³ An increase or decrease in weight was seen in ambient sample as well as control, thus no effect was attributed to contaminants.

4.1.2.3 Toxicity Identification Evaluations

Investigation of the causes of toxicity using TIE methods proved difficult due to generally low acute toxicity of water samples, and the confounding factors due to high salinity effects on *H. azteca*. Salinity affected the chemistry of water samples in a way that reduced the effectiveness of some of the TIE manipulations, in particular the addition of STS to bind metals, and addition of esterase and bovine serum albumin (BSA) in efforts to identify pyrethroid toxicity. Below we list and describe the results of TIEs conducted in 2006-2007.

Site 323 (7/12/2006): Toxicity to *H. azteca* was observed at site 323, collected on July 12, 2006. Relative survival was significantly reduced to 50% by day six of the test. PBO did not enhance acute toxicity, but in fact reduced it, and the same pattern was seen for the “high salinity control”. A Phase I Toxicity Identification Evaluation (TIE) was initiated. Salinity of the water was 15.3 ppt, which is close to the tolerance limit for this species. We therefore tested a series of salinities to evaluate if salinity was the cause of reduced survival. The results are shown in Table A29 (Appendix A). Organic chemicals (eluate addback treatment) as well as high salinity were likely contributing factors in the observed toxic effects.

Site 711 (4/12/2007): Toxicity (47% mortality within 10 days) to *H. azteca* was observed at site 711, in a sample collected on April 12, 2007. Although the toxicity was below the trigger for TIE testing (50% mortality within 7 days), an attempt was made to identify the toxicant in this sample, and a Phase I Toxicity Identification Evaluation (TIE) was initiated. The results of the TIE are shown in Table A69-1 (Appendix A). Toxicity in the original sample was lost by the time the TIE could be completed (90% survival), and the cause of toxicity could not be determined. The metal chelators EDTA and STS did not reduce toxicity. STS by itself appears to be toxic to *H. azteca*. Addition of esterase also introduced toxicity. It is possible that enzyme break-down led to toxic components. These two compounds continued to present problems in TIEs with *H. azteca*, and will not be used in future work.

Site 405 (8/8/2007): Survival of *H. azteca* was significantly decreased after PBO addition to the ambient sample. Although this effect was also seen in the respective high salinity control, a TIE focused on the identification of pyrethroids was initiated. These treatments include extracting organic chemicals using a C8 column and testing the concentrated column eluate, and testing the ambient sample at reduced temperature, with the addition of PBO, esterase, and BSA. Results are shown in Table A86-1 (Appendix A). The C8 eluate (concentrated 3-fold) was more toxic than the respective solvent (MeOH) control, indicating that organic chemicals contributed to the toxicity. Pyrethroid insecticides likely caused at least part of the toxicity, since low temperature increased toxicity in this sample from (77% to 47% survival), and PBO enhanced toxicity. However, analytical chemistry did not detect pyrethroid insecticides (Table 13, below). High salinity likely contributed significantly to the toxicity seen in this sample.

Site 405 (9/4/07): Survival of *H. azteca* was significantly decreased in an ambient sample (13%) collected on October 2, 2007 as well as in the respective PBO treatment (15%) compared to the high EC control. The respective high EC control also showed reduced survival (38%) indicating that high salinity was contributing to the high mortality, but an additional stressor was present. Although it is very difficult to separate a contaminant signal from a high salinity signal,

a TIE was initiated on September 12, 2007. Results are shown in Table A91-1 (Appendix A). The majority of the toxicity in the original, ambient sample was no longer detectable, making the interpretation of TIE results difficult. The C8 eluate (concentrated 3-fold) was more toxic than the respective solvent (MeOH) control, indicating that organic chemicals contributed to the toxicity. Both metal chelators, EDTA and STS, caused toxicity in high salinity laboratory control water, but not in the ambient sample. In order to appropriately interpret these results, further investigations on the interactions of metal chelators with high EC water are needed.

Site Hood (10/2/07): Survival of *H. azteca* was significantly decreased (43%) in a sample collected on October 2, 2007. Addition of PBO alleviated toxicity by a factor of 2, suggesting that organophosphate insecticides caused the observed toxicity. Chemical analysis resulted in no detectable concentrations of organophosphate insecticides. Although the toxicity was below the threshold triggering a TIE (50% mortality by day 7), a TIE was initiated on October 21, 2007. Results are shown in Table A96-1 (Appendix A). Toxicity was no longer present in the original ambient water sample, and therefore the chemical toxicant group could not be further identified. However, the initial signal obtained by the addition of PBO is strong evidence for OP insecticides. The fact that the signal disappeared within 3 weeks, and OPs were not detected by chemical analysis, indicates that the toxicity may have been due to a mixture of chemicals with a similar mechanism of action as OPs.

4.1.2.4 Analytical Chemistry

Water samples submitted for chemical analysis showed noticeable effects on one or more bioassay endpoints: survival, survival after PBO addition, growth or growth after PBO addition. Results from chemical analyses of water samples obtained to date are shown in Table 13. Nine field samples analyzed during the reporting period contained detectable concentrations of pesticides: A sample from site 340 caused a significant reduction in *H. azteca* survival after PBO addition (Table 11 a), and contained 3 ng/L cyfluthrin and 16 ng/L esfenvalerate. Two samples from site 405 caused significant mortality (9/4/07, 10/4/07) and contained 3 ng/L esfenvalerate, and 5 ng/L permethrin, respectively.

Several samples that caused a significant reduction in *H. azteca* growth contained detectable amounts of pyrethroid pesticides: Site 902 sampled on 8/22/06 contained 5 ng/L cyfluthrin and 24 ng/L permethrin; site 340 sampled 2/13/07 contained 63 ng/L cyfluthrin, and sites 915 and 508 sampled on 2/28/07 and 3/1/07, respectively, contained 2 and 3 ng/L lambda-cyhalothrin. A sample from Light 55 collected 2/1/07 contained 6 ng/L diazinon.

Other stressors were likely affecting *H. azteca* in some of these samples. For example, the presence of 5 ng/L esfenvalerate at site 405 (9/4/07) would be unlikely to cause >85% mortality. Contrary to that, the amount of suspended material may alleviate toxicity due to contaminants, in particular the hydrophobic pyrethroids. For example, a concentration of 63 ng/L cyfluthrin detected at site 340 on 2/13/07 would be expected to cause significant mortality, but in this case resulted in only a growth reduction after PBO addition. Further studies to trace the fate of pyrethroid insecticides during sampling and testing are scheduled.

Fate of Pyrethroid Insecticide during Sampling and Testing: As of June 2007, water samples for chemical analysis were preserved by addition of the solvent DCM due to concerns that toxic chemicals, in particular pyrethroid insecticides, could break down during storage. The comparative analysis of a sample from site 405 (10/4/2007) spiked with DCM and without DCM shows that this concern was justified. The DCM-spiked sample yielded a detectable concentration of esfenvalerate, while the non-spiked sample resulted in no detection (Table 13). While samples taken for chemical analysis are stored in glass bottles at 4°C, water samples tested for toxicity are sampled in plastic cubitainers (for safety reasons). To evaluate how this process would affect bioassay and analytical results, we prepared a “mock” spiked sample (sample 409 - Pacheco Creek), Table 13, Appendix A: Table A97) containing 26.5 ng/L permethrin, took one subsample for chemical analysis (no DCM) and stored it until bioassay results were available (approx. 14 d), then sent the sample for analysis of pyrethroids. Only 10 ng/L permethrin was detected, about one third of the original nominal concentration.

Sorption data were collected by Michelle Hladik at the USGS in Sacramento, CA (funding provided by US EPA IAG# DW-14-92230901-0). A mixture of 14 pyrethroids (400 ng/L) was spiked into American River water and filled into plastic 1-gallon and 5-gallon cubitainers (3 replicates each size) used for sampling. Cubitainers were allowed to sit for seven days in the dark at 4°C (1-gal) or room temperature (5-gal). UCD-ATL stores all samples at 4°C. After seven days the containers were agitated for at least one minute and then the water was poured out. The containers were rinsed with methanol to remove the remaining pyrethroids. The results of chemical analysis showed that the percentage of pyrethroid adsorbed to container walls was pyrethroid-specific and higher in the small cubitainers, with 0% (tetramethrin) to 7.0 (cyfluthrin)% of the pyrethroids adsorbed to the 1-gal cubitainers, and 0% (allethrin, tetramethrin) to 3.3% (cyhalothrin) adsorbed to the 5-gal cubitainers.

Table 13. Results of analytical chemistry on water samples that caused significant changes in *H. azteca* survival or growth.

Site ID	Collection Date	Scan Type	Results
323	6/14/2006	metal, OP scan	54 µg/L barium, 75 µg/L zinc
405	6/28/2006	OP scan	ND
812	6/29/2006	pyrethroid scan	ND
711	8/22/2006	pyrethroid scan	ND
Light 55	8/22/2006	pyrethroid scan	ND
902	8/22/2006	pyrethroid scan	0.005 µg/L cyfluthrin, 0.024 µg/L permethrin
609	8/23/2006	pyrethroid scan	ND
508	9/21/2006	pyrethroid scan	ND
504	9/21/2006	pyrethroid scan	ND
405	9/21/2006	pyrethroid scan	ND
902	10/3/2006	pyrethroid scan	ND
910	10/3/2006	pyrethroid scan	ND
711	10/3/2006	pyrethroid scan	ND
504	1/16/2007	pyrethroid scan	ND
910	2/1/2007	pyrethroid scan	ND

Table 13, continued

915	2/1/2007	pyrethroid scan	ND
Light 55	2/1/2007	OP, carbamate scan	0.006 µg/L diazinon
340	2/13/2007	pyrethroid scan	0.063 µg/L cyfluthrin
504	2/14/2007	pyrethroid scan	ND
902	2/15/2007	pyrethroid scan	ND
Light 55	2/15/2007	pyrethroid scan	ND
915	2/28/2007	pyrethroid scan	0.002 µg/L lambda cyhalothrin
711	2/28/2007	dissolved metals scan	0.60 mg/L boron, 100 mg/L calcium, 30 mg/L magnesium, 20 mg/L silicon, 100 mg/L sodium
508	3/1/2007	pyrethroid scan	0.003 µg/L lambda cyhalothrin
602	3/14/2007	pyrethroid scan	ND
609	3/14/2007	pyrethroid scan	ND
704	3/29/2007	OP scan	ND
711	4/12/2007	OP scan	ND
711	5/22/2007	OP, pyrethroid scan	ND
508	5/23/2007	pyrethroid scan	ND
902	6/6/2007	OP scan	ND
812	6/6/2007	OP scan	ND
910	6/6/2007	OP scan	ND
711	6/20/2007	OP scan	ND
405	7/10/2007	comprehensive organics/inorganics scan	ND
804	7/10/2007	comprehensive organics/inorganics scan	0.047 ug/L piperonyl butoxide
602	7/10/2007	comprehensive organics/inorganics scan	ND
340	7/25/2007	pyrethroid scan	0.003 ug/L cyfluthrin 0.016 ug/L esfenvalerate
405	8/8/2007	pyrethroid scan	ND
405	8/22/2007	extracted and hold	results not received
405	9/4/2007	comprehensive org. scan	0.003 µg/L esfenvalerate
405	9/19/2007	extracted and hold	results not received
602	9/19/2007	pyrethroid scan	ND
409*	10/5/2007	pyrethroid scan	0.010 µg/L permethrin
Hood	10/2/2007	OP scan	ND
405	10/4/2007	pyrethroid scan	ND
405 with DCM	10/4/2007	pyrethroid scan	0.005 µg/L permethrin
Hood	10/16/2007	pyrethroid scan	ND
Hood	10/30/2007	OP scan	ND
Light 55	10/31/2007	OP scan	ND
711	10/31/2007	OP & pyrethroid scan	ND/ND

ND - Analyte not detected at or above the reporting limit.

*Mock sample: Non-toxic delta water (sites 405/609; salinity adjusted to 27 mS/cm) spiked with 26.5 ng/L permethrin, not preserved with DCM.

4.1.2.5 Effect of Salinity on *H. azteca* Survival and Growth

We analyzed data from control treatments of tests conducted during 2006-2007 with *H. azteca* to determine salinity-specific effects and discriminate between those and other site-specific factors that affected amphipod survival (Figures 3, 4). We also compared treatments with and without PBO to determine if PBO addition would negatively affect test animals in combination with salinity. MANOVA analyses of high EC control survival and weight data showed that PBO treatment did not affect the regressions of survival or weight on EC (Survival: PBO effect: $F_{1,84} = 0.0001$, $P = 0.925$, PBO*EC Interaction: $F_{1,84} = 0.0005$, $P = 0.8419$, Weight: PBO effect: $F_{1,53} = 0.0038$, $P = 0.6551$, PBO*EC Interaction: $F_{1,53} = 0.0031$, $P = 0.6861$). In these MANOVAs, performance (survival or weight) of untreated high EC control animals and performance of PBO-treated high EC control animals were the response variables and Log EC was the independent variable.

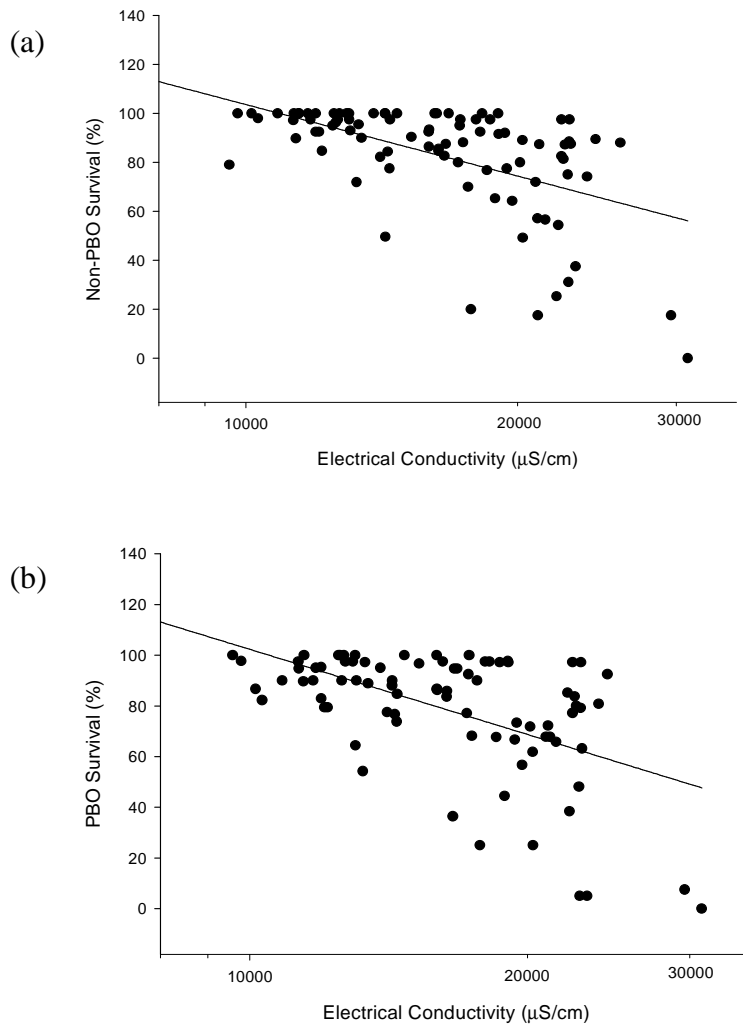


Figure 3. Relationships between survival and EC in high conductivity control treatments in a *H. azteca* 10-day tests, in (a) ambient samples and (b) samples treated with PBO (linear regressions, non-PBO: $N = 92$, adjusted $r^2 = 0.298$, $P < 0.0001$, PBO: $N = 86$, adjusted $r^2 = 0.241$, $P < 0.0001$).

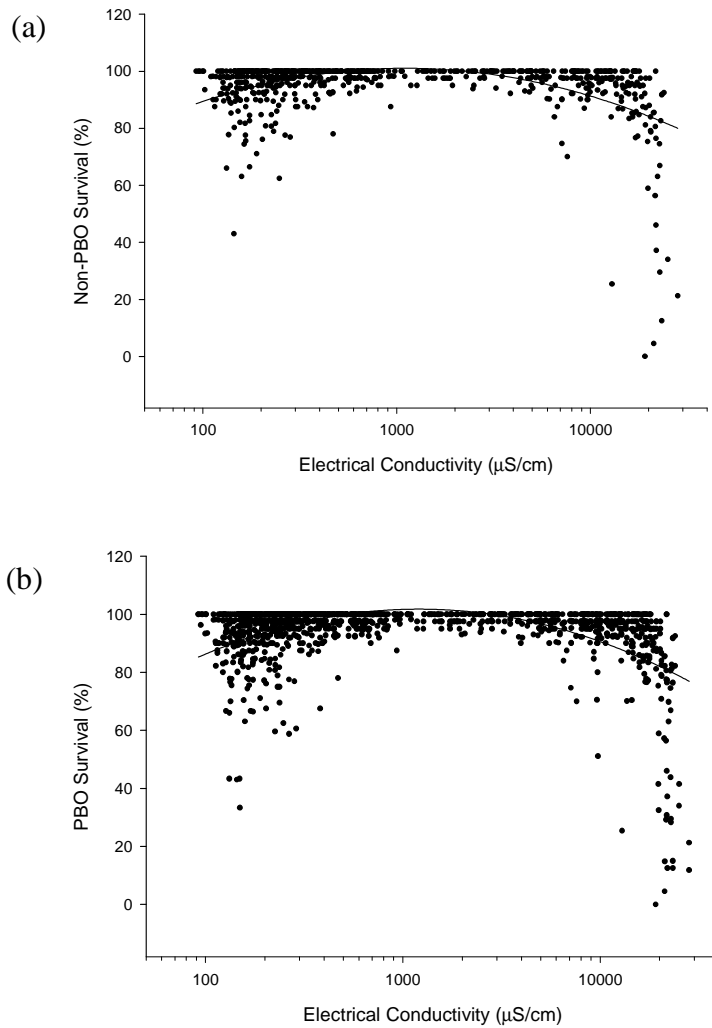


Figure 4. Relationships between survival and EC in ambient delta water samples in a *H. azteca* 10-day chronic toxicity test, in (a) ambient samples and (b) samples treated with PBO (polynomial regressions, non-PBO: $N = 704$, adjusted $r^2 = 0.188$, $P < 0.0001$, PBO: $N = 675$, adjusted $r^2 = 0.212$, $P < 0.0001$).

Site and Seasonal differences in *H. azteca* growth: The parabolic curve fits of EC to weight data (Figures 5, 6) were used in ANCOVA models aimed at revealing any sites or seasons where *H. azteca* weights deviated from expectations based on conductivity of the sample water. Few significant deviations were found, and those tended to occur at the low and high extremes of the conductivity spectrum. This indicates that the deviations occurred because the parabolic curve fit to the EC effect may not adequately describe the effects of EC at very low and high conductivities. No strong evidence was found that would suggest that major site to site or seasonal differences in *H. azteca* weight were present that could not be accounted for by differences in conductivity. It should be noted, as our weight MSD readings show, that detecting

small to moderate differences between sites and seasons is challenging given the low statistical sensitivity of comparisons involving weight, and many potential differences between sites may not be revealed by this analysis.

Season-specific analysis of growth data revealed trends in growth deviations from values expected based on EC at each sites. Figures in Appendix B show *H. azteca* weight by season and site. The ANCOVA models show that the differences in *H. azteca* weights among sites may be largely explained by the effects of conductivity, but model coefficients for the effects associated with specific sites are provided to indicate potential between-site effects that were not adequately explained by the effects of conductivity. Sites 711, 405 and 602 tended to have lower than expected growth, while site 704, 804, 902, 915 had instances of higher than expected growth.

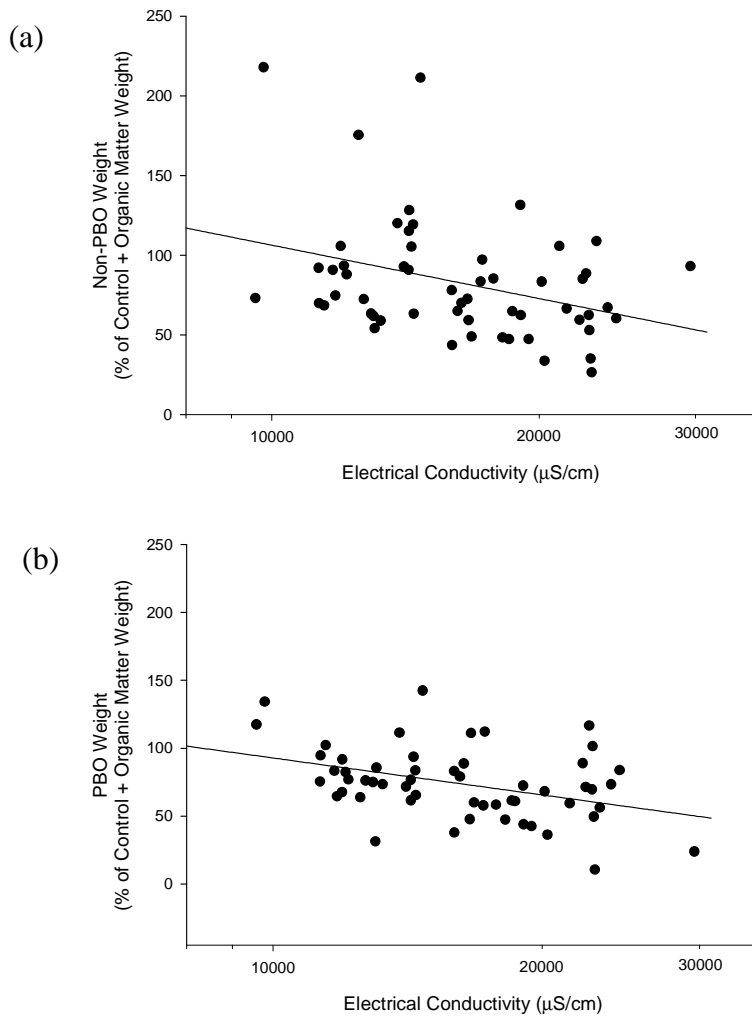


Figure 5. Relationships between weight and EC in high conductivity control waters in a *H. azteca* 10-day chronic toxicity test, in (a) ambient samples and (b) samples treated with PBO (linear regressions, non-PBO: $N = 55$, adjusted $r^2 = 0.091$, $P = 0.014$, PBO: $N = 55$, adjusted $r^2 = 0.144$, $P < 0.0025$).

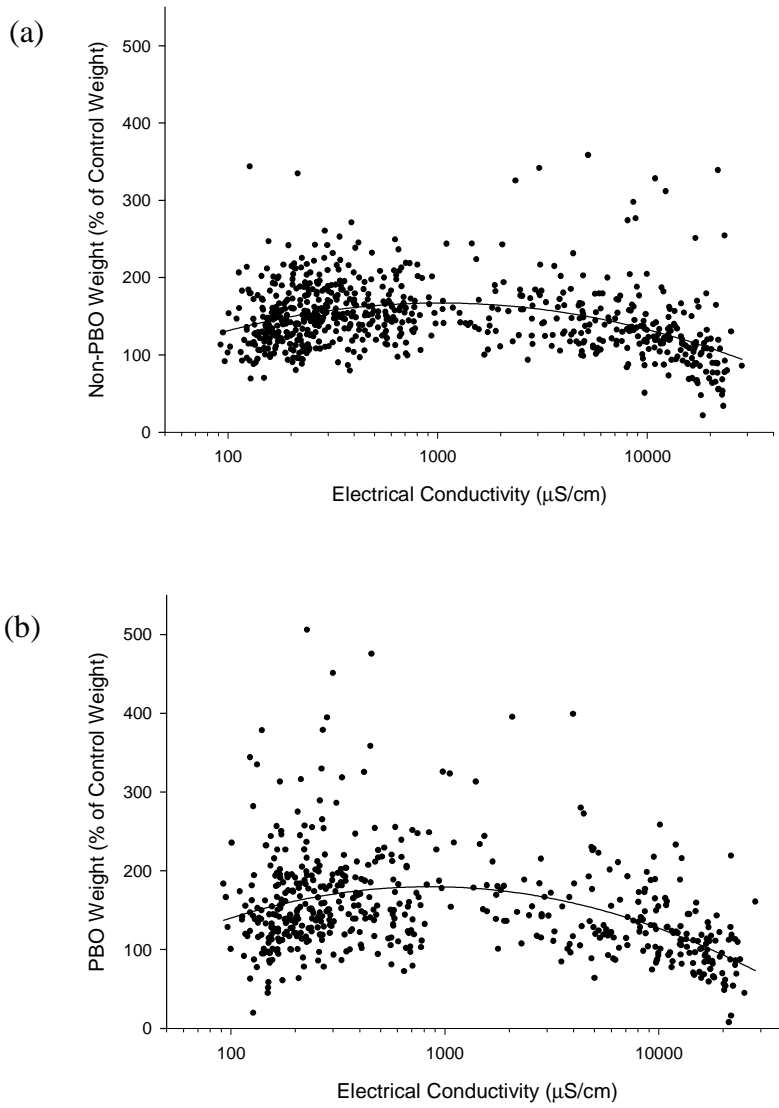


Figure 6. Relationships between weight and EC in ambient delta water samples in a *H. azteca* 10-day chronic toxicity test, in (a) ambient samples and (b) samples treated with PBO (polynomial regressions, non-PBO: $N = 702$, adjusted $r^2 = 0.133$, $P < 0.0001$, PBO: $N = 540$, adjusted $r^2 = 0.153$, $P < 0.0001$).

4.1.2.6 Effect of Ammonia on *H. azteca* Survival and Growth

Regression models that controlled for the effects of site-specific EC differences by parabolic curve fits (see above) were used to detect possible effects of ammonia on *H. azteca* survival and weight. Data were analyzed together at all sites over the two year study period, and also separately at each site and during each season. Overall, ammonia had significant effects on *H. azteca* weight both when measured as ammonia nitrogen and when measured as unionized ammonia, but no significant effect on *H. azteca* survival was observed (Table 14 a). When analyzed by site, total ammonia-N concentrations were positively related to survival at sites 504, 609 and 804, and negatively related to survival at Light 55 (Table 14 b). Ammonia-N and/or unionized ammonia concentrations were negatively related to *H. azteca* growth at sites 323, 812 and Light 55 (Table 14 d). Ammonia nitrogen and unionized ammonia measurements gave essentially parallel results, although unionized ammonia revealed relationships with amphipod growth at sites 323 and Light 55 that did not appear in the analysis on ammonia-N. A similar analysis of ammonia effects on survival and weight during different seasons found only one significant association: survival during the winter of 2007 was negatively associated with levels of ammonia-N and unionized ammonia (Table 14 c). Similarly, amphipod growth was negatively associated with unionized ammonia during the same period (Table 14 e).

Table 14 a. Magnitude and significance of ammonia effects on the survival and weight of *H. azteca* exposed to ambient Delta waters not treated with PBO in 10-day chronic water column toxicity tests. Ammonia effects were measured in regression models controlling for the effects of EC differences by parabolic curve fits.

Response	Ammonia Nitrogen Effect			Unionized Ammonia Effect		
	<i>N</i>	Coeff. ¹	<i>P</i>	<i>N</i>	Coeff. ¹	<i>P</i>
Survival (%)	702	0.1	0.9338	702	0.1	0.9086
Weight (% of control)	702	-13.4	0.0021	702	-20.4	0.0002

1. Positive coefficients indicate positive correlations, negative coefficients indicate negative correlations.

Table 14 b. Magnitude and significance of ammonia effects on site-specific survival (% of control) of *H. azteca* exposed to ambient Delta waters (without PBO) in 10-day chronic water column toxicity tests. Ammonia effects were measured in regression models controlling for the effects of EC differences by parabolic curve fits.

Site	N	Ammonia Nitrogen Effect		Unionized Ammonia Effect	
		Coeff. ¹	P	Coeff. ¹	P
323	14	41.3	0.4854	11.8	0.898
340	38	-6.49	0.1183	-5.4	0.4004
405	47	14.9	0.2319	6	0.7214
504	50	6.5	0.0048	12.4	0.0001
508	50	1.31	0.5201	3.4	0.1717
602	49	7.2	0.2189	14.2	0.0621
609	50	7.6	0.0012	9.7	0.0027
704	50	2.1	0.0937	2.9	0.0682
711	50	-8.7	0.1298	-6.34	0.2057
804	50	3.1	0.0106	3.8	0.0136
812	48	-0.7	0.8361	-3	0.4149
902	50	-0.5	0.8292	0.5	0.8457
910	50	1	0.4619	1.1	0.5057
915	50	1.9	0.2625	2.6	0.2368
Hood	8	11.7	0.7186	6.6	0.8351
Light 55	48	-5.3	0.0344	-5.6	0.045

¹ Positive coefficients indicate positive correlations, negative coefficients indicate negative correlations.

Table 14 c. Magnitude and significance of ammonia effects by season on the survival of *H. azteca* exposed to ambient Delta waters not treated with PBO in 10-day chronic water column toxicity tests. Ammonia effects were measured in regression models controlling for the effects of EC differences by parabolic curve fits.

Season	Ammonia Effect			Unionized Ammonia Effect		
	N	Coeff. ¹	P	N	Coeff. ¹	P
Jan – Mar 2006	82	1.6	0.1651	82	1.3	0.3153
Apr – Jun 2006	105	3.3	0.1992	105	3.5	0.1895
Jul – Sep 2006	86	-0.1	0.9788	86	1.2	0.7870
Oct – Dec 2006	84	1.4	0.4109	84	1.0	0.6462
Jan – Mar 2007	98	-3.4	0.0134	98	-4.6	0.0040
Apr – Jun 2007	86	-3.2	0.0764	86	-2.2	0.3464
Jul – Sep 2007	81	-5.3	0.3481	81	-6.6	0.4010
Oct – Dec 2007	82	0.9	0.8437	82	3.5	0.5012

¹ Positive coefficients indicate positive correlations, negative coefficients indicate negative correlations.

Table 14 d. Magnitude and significance of ammonia effects on site-specific growth (% of control) of *H. azteca* exposed to ambient Delta waters (without PBO) in 10-day chronic water column toxicity tests. Ammonia effects were measured in regression models controlling for the effects of EC differences by parabolic curve fits.

Site	Ammonia Nitrogen Effect			Unionized Ammonia Effect		
	N	Coeff. ¹	P	N	Coeff. ¹	P
323	14	-45.3	0.3668	14	-141.3	0.0485
340	38	-19.88	0.1743	38	-23	0.3095
405	47	-23.9	0.3071	47	-24	0.4466
504	50	-36.6	0.0728	50	-49.8	0.0809
508	50	-4.6	0.8117	50	-1.3	0.9545
602	49	-30.8	0.1634	49	-19	0.515
609	50	-8.6	0.6961	50	-15.9	0.5988
704	50	-21.8	0.3082	50	-32.9	0.2269
711	50	4.7	0.8623	50	-34.9	0.1321
804	50	2.3	0.8999	50	0.6	0.9783
812	48	-68.3	0.0048	48	-73.6	0.0089
902	50	-15.6	0.2458	50	-14	0.38
910	50	13.7	0.394	50	7.2	0.6944
915	50	-2.1	0.8802	50	-10.4	0.5538
Hood	8	-88.1	0.0657	8	-48.7	0.3782
Light 55	48	-21.4	0.134	48	-35.2	0.0248

¹ Positive coefficients indicate positive correlations, negative coefficients indicate negative correlations.

Table 14 e. Magnitude and significance of ammonia effects by season on the weight (as % control) of *H. azteca* exposed to ambient Delta waters not treated with PBO in 10-day chronic water column toxicity tests. Ammonia effects were measured in regression models controlling for the effects of EC differences by parabolic curve fits.

Season	Ammonia Nitrogen Effect			Unionized Ammonia Effect		
	N	Coeff. ¹	P	N	Coeff. ¹	P
Jan – Mar 2006	82	-0.8	0.9488	82	7.5	0.6159
Apr – Jun 2006	105	-25.6	0.0697	105	-24.7	0.0974
Jul – Sep 2006	86	-5.5	0.5838	86	-11.3	0.3987
Oct – Dec 2006	84	-3.2	0.7195	84	-18.5	0.0868
Jan – Mar 2007	98	-13.5	0.1560	98	-23.7	0.0306
Apr – Jun 2007	86	-4.9	0.7768	86	6.9	0.7646
Jul – Sep 2007	81	20.4	0.3773	81	13.1	0.6861
Oct – Dec 2007	82	-18.9	0.1555	82	-12.6	0.4103

1. Positive coefficients indicate positive correlations, negative coefficients indicate negative correlations.

4.2 Laboratory Experiments

Pyrethroid Toxicity at Environmentally Relevant Concentrations: Lethal and Sublethal Effects in the Amphipod *Hyaella azteca* Susanne M Brander*, Inge Werner, Linda A Deanovic;

Introduction

Pyrethroid pesticide use during 2000-2003 in the Central Valley of California (San Joaquin & Sacramento) was doubled from 1990 levels, mainly due to the phasing out of the more toxic OPs for both agricultural and residential applications (Epstein et al. 2000; Oros and Werner 2005). However, over the past decade it has been discovered that while pyrethroids are not acutely toxic to mammals, they are very toxic to fish and aquatic invertebrates (Oros and Werner 2005). This is due to a combination of factors, including the similar physiology of aquatic invertebrates to insects and the potential for disruption of osmoregulation in fish (Clark and Matsumura 1982; Oros and Werner 2005). Pyrethroids disrupt the nervous system by binding to and prolonging the opening of voltage-dependent ion channels, mainly those controlling the passage of sodium, but sometimes chloride and calcium channels as well (Burr and Ray 2004; Marshalonis et al. 2006; Shafer and Meyer 2004). Because the opening of these channels controls the firing of neurons, the consequence of extended opening is convulsions, paralysis and eventually death (Shafer & Meyer 2004; Oros & Werner 2005).

Pyrethroids are highly lipophilic and tend to bind to sediments, and therefore it has been argued that this decreases their toxicity substantially (Leahey 1985). However, these compounds can remain in the water column for days to weeks and are soluble enough to render biological harm to vulnerable organisms, especially considering that pyrethroids are toxic in the ppb range (Oros & Werner 2005). Due to their unique chemical properties, pyrethroids may be harmful to both pelagic and benthic species. *Hyaella azteca*, an epibenthic organism prevalent in the Sacramento / San Joaquin Delta which receives run-off from the CA Central Valley, may be exposed to these pesticides via both routes and has already been found to be highly sensitive to sediment-bound pyrethroids (Weston et al. 2005; Weston et al. 2004). Hence it was chosen for this study.

Permethrin and cyfluthrin, two pyrethroid pesticides found in the SSJ Delta, are toxic to *Hyaella azteca* at the ppb range, well within levels measured in the region (Amweg et al. 2005; Amweg et al. 2006b). In recent studies, sediment-bound cyfluthrin and permethrin had LC50s as low as 12.5 ng/g (ppb) and 57 ng/g, respectively, in *Hyaella* (Amweg et al. 2005; Amweg et al. 2006b; Weston et al. 2004). Permethrin toxicity has been observed at the ppb level in other crustaceans, fish and amphibians as well (DeLorenzo et al. 2006; Oros and Werner 2005).

Newer generation “type II” pyrethroids, which degrade more slowly, bind more effectively to sodium channels and therefore prolong firing longer than older “type I” pyrethroids. This results in higher toxicity at lower concentrations (Leahey 1985). Of the top

five pyrethroids in use in this region, permethrin (type I) is the most frequently used and least toxic, and cyfluthrin (type II) is the fifth most used but ranks second in toxicity (Oros & Werner 2005). As a result of the mechanistic enhancements endowed upon type II pyrethroids (such as cyfluthrin), they may have a toxic potency up to 20-fold that of a type I pyrethroid like permethrin (Oros and Werner 2005).

The toxicity of both type I and II pyrethroids is further amplified by the pesticide synergist piperonyl butoxide (PBO), which can increase the toxicity of pyrethroids 10 to 150-fold, depending on the formulation (Wheelock et al. 2004), through inhibition of the enzymes that metabolically deactivate the pyrethroid molecules (Amweg et al. 2006a). In addition to the threat posed by PBO, classes of pesticides commonly found together in aquatic ecosystems that have different targets, such as pyrethroids and organophosphates, or pyrethroids and carbamates, have been found to be synergistic (Corbel et al. 2004; Denton et al. 2003). However, little is known about the combined toxicity of specific type I and II pyrethroids.

The objective of this study was to use a local, sensitive species to evaluate the toxicity of environmentally relevant concentrations and mixtures of two pyrethroid pesticides detected in the water column of the SSJ Delta, in Old River at the mouth of Holland Cut (Figure 1). Although a number of studies have utilized *Hyaella azteca* to examine the toxicity of pyrethroids bound to sediments (Weston et al. 2004; Weston et al. 2005; Amweg et al. 2005; Amweg et al. 2006), this is one of the first studies to evaluate the combined toxicity of permethrin (type I) and cyfluthrin (type II) to *H. azteca* in the water column at levels measured in the SSJ Delta water column.

Methods

A water sample collected on August 22, 2006 at Site 902 (Old River at the mouth of Holland Cut, 38-01-09.1N, 121-34-55.9W) caused a significant reduction (52%) of *H. azteca* growth after PBO addition. (Werner I., unpublished data). Chemical analysis of whole water samples revealed the presence of two pyrethroid pesticides: 0.005 µg/L (ppb) cyfluthrin, and 0.024 µg/L (ppb) permethrin. To verify if these compounds could be responsible for the observed toxic effects, a laboratory experiment was performed in 2007 and repeated in 2008.

Cyfluthrin (Baythroid™, 98% mix of isomers; Figure 1) and permethrin (31.8% cis, 67.4% trans; Figure 2) were purchased from Chem Service, Inc. in West Chester, PA. Stock solutions were made in methanol and cyfluthrin and permethrin stocks were spiked into laboratory control water consisting of deionized water amended to US EPA moderately hard standards (DIEPAMHR) to yield the following nominal concentrations:

Cyfluthrin: 0.0025, 0.005, 0.01 µg/L (ppb)

Permethrin: 0.012, 0.024, 0.048 µg/L (ppb)

Cyfluthrin + Permethrin: 0.0025 + 0.012, 0.005+0.024, 0.01+0.048 µg/L

Confirmatory chemistry was performed at the California Department of Fish & Game Laboratory in Sacramento, CA. Nominal and measured concentrations can be found in Table 1.

Figure 1. Chemical structure of permethrin

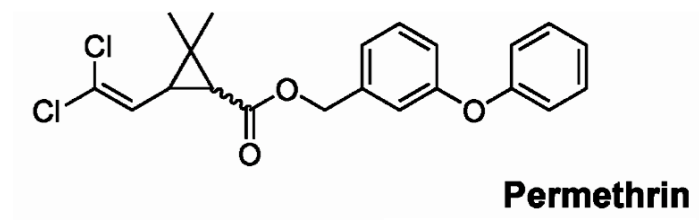


Figure 2. Chemical Structure of Cyfluthrin .

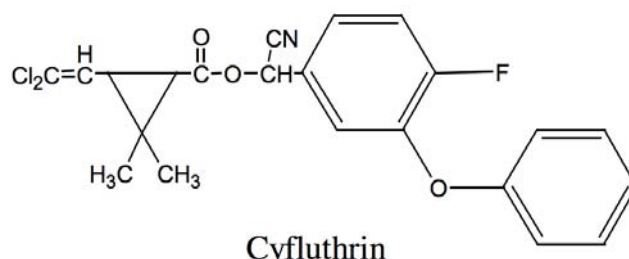


Table 1.
Nominal and Measured Concentrations

treatment	nominal concentration (ppb)	measured concentration (ppb)	
		2007	2008*
cyfluthrin (1/2 DL)	0.0025	0.0029	0.002
cyfluthrin (DL)	0.005	0.0051	0.003
cyfluthrin (2 x DL)	0.010	0.0104	0.004
permethrin (1/2 DL)	0.012	0.0119	0.004
permethrin (DL)	0.024	0.0254	0.008
permethrin (2 x DL)	0.048	0.0573	0.016

DL = detected level

*2008 nominal concentrations instead of 2008 measured concentrations for cyfluthrin were used in the statistical analysis, as mortality levels indicate that significant degradation occurred in the samples sent to DFG for extraction. Confirmation of this issue is pending.

Tests were conducted with and without piperonyl butoxide (PBO) addition - a commonly used pesticide synergist. A five parts per million (5 ppm) stock solution of PBO was prepared and added to 400 ml of water sample for a final concentration of 25 parts per billion (ppb).

In both 2007 and 2008, biological testing conducted in the Aquatic Toxicology Laboratory (ATL) at the University of California, Davis (UCD) adhered to EPA protocol for a

10-day chronic exposure using *H. azteca* (USEPA 1994). *H. azteca* were purchased from Aquatic Research Organisms (New Hampshire, MD). The 10-day tests consist of five replicate 250 ml glass beakers each containing 100 ml of sample, a one-square-inch piece of nitex screen (a substrate for the *H. azteca* to cling to), and 10 organisms. Tests were initiated with 7 to 14 day old *H. azteca*. Animals in each replicate were fed 1000 l of YCT (a mixture of yeast, organic alfalfa and trout chow) on test initiation and days 2, 4, 6, 8, as well as on day 5, when 75% of the test water was renewed. Each series of tests included a standard laboratory control and a solvent (0.025% MeOH) control. Tests were conducted in a $23 \pm 2^\circ$ C water bath with a 16h:8h L:D photoperiod. Mortality was recorded daily. On day 10, half of the surviving *H. azteca* were dried and weighed to determine dry tissue weight/individual and relative growth. The remaining animals were flash-frozen in liquid nitrogen and stored for biochemical analysis.

After 2007 testing only, a Bradford protein analysis was performed on *H. azteca* that were still alive at test termination. Bovine serum albumin (BSA) was used as a control. Briefly, amphipod samples were homogenized on ice in a hypotonic solution containing 66 mM Tris-HCl (pH 7.5), 0.1% Nonidet, 10 mM EDTA, 10 mM DTT and protease inhibitors. Following centrifugation at 4°C, supernatants were collected, and total protein concentration was determined using the Biorad DC Protein Assay based on Lowry et al. (1951).

Statistical Analysis: We analyzed survival data using logistic regression. Regression models were as follows: $Mortality = \exp(bX)/(1+\exp(bX)) + \Sigma$, where *b* is a vector of parameters, *X* is a matrix of predictor variables, and Σ is a binomial error term. We considered univariate models containing terms for cyfluthrin concentration, permethrin concentration, and presence of PBO as well as models containing all possible combinations of these terms and their interactions, so we could ascertain whether antagonism or synergism was occurring between cyfluthrin and permethrin. We then used a version of Akaike’s Information Criterion corrected for small sample sizes (AIC_c; Burnham and Anderson 1998) to select the most parsimonious model from among the 21 considered. All regressions were performed in Matlab 7.0 (Mathworks Inc, Natick, MA).

We also calculated the LC50 for each of the individual pesticides in their respective solitary treatments (i.e. cyfluthrin only, permethrin only). Because concentration-based LC50s could not be calculated for the cyfluthrin / permethrin mixture treatments, we used a dilution index to estimate the combined concentrations that would be required to cause a specific proportion mortality. The index is based on setting the values of the actual levels of cyfluthrin and permethrin measured in the SSJ Delta each equal to 1. Calculations for the dilution index are shown in Table 3.

Table 2. Dilution index calculations.

treatment level		dilution index value		index total
cyfluthrin	permethrin	cyfluthrin	permethrin	
0.0025	0.012	0.5	0.5	1
0.0050*	0.024*	1	1	2
0.0100	0.048	2	2	4

*Levels measured at site 902

Results

For the two analyses of mortality as a function of either permethrin or cyfluthrin alone, the most parsimonious models (as identified by AIC_c) were the full models, with terms for the pesticide and the pesticide spiked with PBO. LC50s were calculated based on the percentage mortality in each group of treatments combined from 2007 and 2008 (Figures 4,5). The LC50 for cyfluthrin alone was calculated to be 0.0065 ppb (6.5 parts per trillion), and the LC50 for permethrin alone was estimated to be 0.0465 ppb (46.5 parts per trillion). The addition of 25 ppb PBO resulted in significantly lower LC50s for both cyfluthrin and permethrin, at 0.0033 ppb and 0.0139 ppb, respectively. PBO doubled the toxicity of cyfluthrin and more than tripled the toxicity of permethrin.

For the analysis of mortality using the entire dataset including treatments with both pesticides, model selection using AIC_c did not identify a single most parsimonious regression model, but the best 2 models represented 97.5% of AIC_c weight (Appendix 1), indicating that the best model has an 97.5% chance of being among that set (Burnham and Anderson 1998). Because the full model (containing terms for cyfluthrin, permethrin, PBO, and all possible interactions) was within this best model set and contains all of the terms appearing in the other top models, we used this model as the best predictor of the data (Table 3). This model describes a negative relationship between cyfluthrin and permethrin (coefficient = -4480.41), indicating that slight antagonism (p = 0.0005) is occurring between these two pesticides.

Table 3. Model coefficients, standard errors and p-values

parameter	cyfluthrin			permethrin			cyfluthrin + permethrin		
	coeff.	S.E.	P value	coeff	S.E.	P value	coeff.	S.E.	P value
intercept	-3.41	0.21	0.0000	-3.30	0.18	0.0000	-3.35	0.14	0.0000
cyfluthrin	525.97	36.59	0.0000				522.07	29.40	0.0000
cyf × PBO	517.27	53.94	0.0000				528.24	50.03	0.0000
permethrin				70.99	5.99	0.0000	69.08	5.40	0.0000
per × PBO				166.09	14.20	0.0000	170.72	13.27	0.0000
cyf × per							-4480.41	1289.87	0.0005
cyf × per × PBO							-49.75	12181.94	0.9967

LC50s were calculated based on the percentage mortality in each group of treatments combined from 2007 and 2008 (Figures 3, 4). The LC50 for cyfluthrin alone was calculated to

be 0.0065 ppb (6.5 parts per trillion), and the LC50 for permethrin alone was estimated to be 0.0465 ppb (46.5 parts per trillion). The addition of 25 ppb PBO resulted in significantly lower LC50s for both cyfluthrin and permethrin, at 0.0033 ppb and 0.0139 ppb, respectively. PBO doubled the toxicity of cyfluthrin and more than tripled the toxicity of permethrin.

Figure 3. Cyfluthrin dose-response

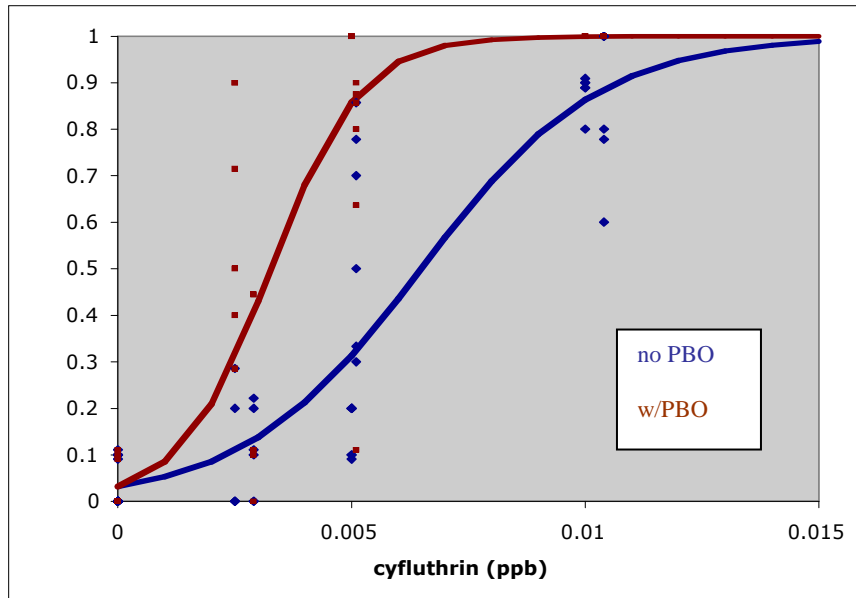
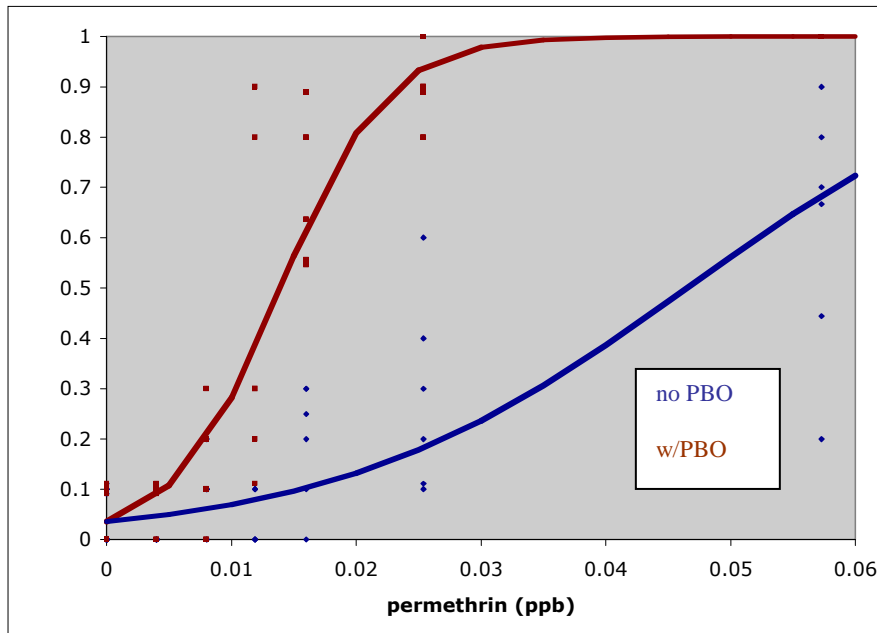
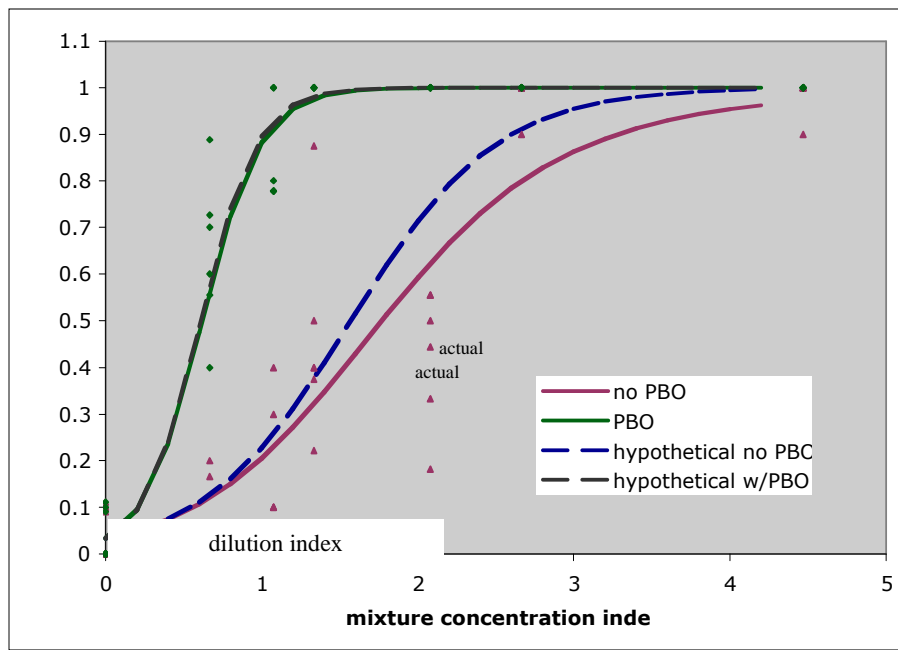


Figure 4. Permethrin dose-response



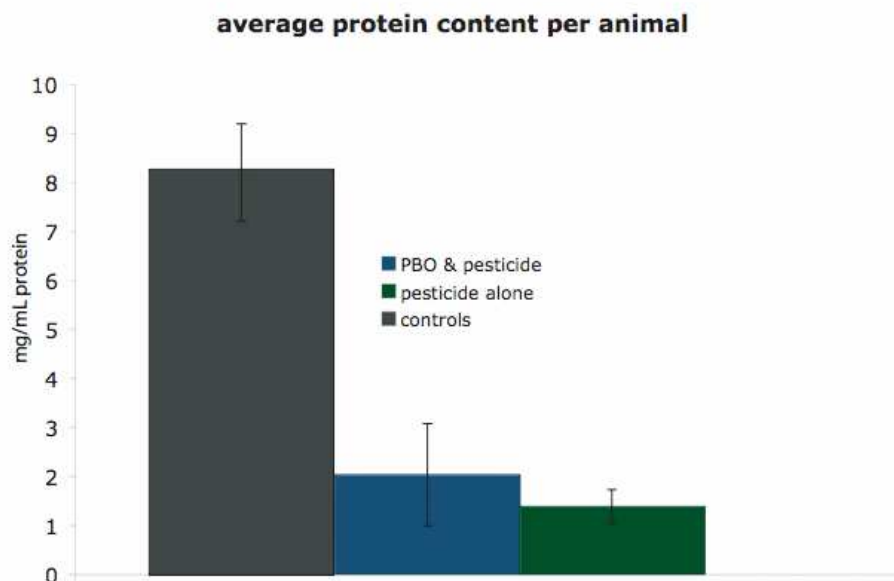
Measuring the toxicity of the permethrin and cyfluthrin mixture was addressed by assigning a dilution index (Table 3) to each treatment level. As shown in Figure 6, 50 % mortality is observed at a dilution index value of approximately 2, which would be equal to a cyfluthrin concentration of 0.005 ppb and a permethrin concentration of 0.012. These results include a negative interaction effect between cyfluthrin and permethrin, which can be seen in the difference between the actual and hypothetical “no PBO” dose-response curves. The “hypothetical” dose-response curves assume an additive interaction, based on a summation of the toxicities of cyfluthrin alone and permethrin alone.

Figure 5. Mixture dose-response: actual vs. hypothetical



The Bradford protein analysis conducted in 2007 showed that amphipods exposed to pyrethroid pesticides or pyrethroids spiked with 25 ppb PBO had significantly less protein than controls ($p < 0.05$) (Figure 6). This precluded any analysis of heat shock proteins as was originally intended, since the protein content of most of the animals that remained alive at the end of the test was too low for HSP analysis. Weight at test termination was also inconclusive in both 2007 and 2008. While pesticide-exposed amphipods weighed less than controls, a dose-response pattern was not evident due to the high variance in weight between replicates.

Figure 6. Bradford Protein Analysis 2007



Discussion

One of the more novel findings of this study is the significant difference in the synergism of permethrin (type I) and cyfluthrin (type II) by PBO. Although a previous study found no difference in the synergism of toxicity by PBO between type I and type II pyrethroids (Wheelock et al. 2004), our results indicate that permethrin toxicity was synergized 3.5 times by PBO, while the toxicity of cyfluthrin was doubled. This effect is present in the data from 2007 and 2008. It may be that because type II pyrethroids are designed to be more resistant to breakdown by P450 enzymes and carboxylesterase to begin with, that inhibiting enzymes that type II pyrethroids are already resistant to does less to increase toxicity than this enzyme inhibition does with type I pyrethroids which are more quickly metabolized to begin with. More simply, the design of type II pyrethroids helps to circumvent the problem with breakage at their ester linkage via carboxylesterase (Leahey 1985) without the addition of PBO.

As expected, the permethrin and cyfluthrin mixture resulted in higher toxicity than either pesticide alone. However, slight but clear antagonism was apparent between the two pesticides. This could be a result of binding site saturation. Cyfluthrin also may be out-competing permethrin for the same binding sites, particularly sodium channel binding sites for which both type I and type II pyrethroids have high affinity for (Leahey 1985, Shafer and Meyer 2004). Cyfluthrin, which breaks down more slowly and is more stable than other pyrethroids, can bind longer than permethrin (Wheelock et al. 2004). For example, perhaps by the time cyfluthrin degrades and permethrin can access the binding site, permethrin has already been metabolized

and is therefore inactive. This could contribute to our observing of a less-than-additive effect. Interestingly, PBO seems to negate any antagonism between cyfluthrin and permethrin. Because both pyrethroids are less resistant to metabolic enzymes in the presence of PBO, perhaps this enhancement overrides any slight antagonism introduced by competition for the same binding sites.

Regardless of whether slight antagonism may be occurring, the single and combined toxicity of permethrin and cyfluthrin at pptr concentrations is cause for concern, as these levels have been detected in the SSJ Estuary and its tributaries (Oros and Werner 2005). And although pyrethroid toxicity may be mitigated by the presence of organic material or fine-grained sediment (Yang et al. 2006), it is clear from the water samples on which this study was based that concentrations high enough to elicit an LC50 level response are periodically present in the water column. Considering the number of other pyrethroids in use in the Central Valley, some of which are more soluble in water and the potential for synergistic interactions with other pesticides (Corbel et al. 2004), and/or residual PBO present in the water column (Amweg et al. 2006a), this is cause for concern.

Heat shock proteins, which are a valuable biomarker in amphipods (Werner and Nagel 1997), should be measured in future studies to examine the sublethal effects of pyrethroids and pyrethroid mixtures. This was one of the original intentions of this study, however, the small size of *H. azteca* results in the requirement of a sample size at least double the size used for this analysis, especially considering the greater than expected mortality and the large reduction in protein content observed in animals exposed to pyrethroids (Figure 6). Further studies using sublethal concentrations should be performed to evaluate the capacity of single pyrethroids and pyrethroid mixtures to cause disruption of cellular homeostasis and other sublethal effects, such as immunotoxicity or endocrine disruption, at part per trillion concentrations. In addition, studies examining the interactions between three or more pyrethroid pesticide mixtures should be conducted, as these types of treatments would more closely mimic conditions in the wild.

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5. Tests with Striped Bass (*Morone saxatilis*)

5.1 Methods

5.1.1 Toxicity Testing

To date, only an initial pilot test and one test with ambient samples from Delta sites have been performed with larval striped bass due to the difficulties in obtaining larvae of this particular strain of striped bass. Two tests with juvenile (80-90 d old) fish were conducted, one in 2005 and one in 2006. We included the 2005 tests in this report, because biomarker data for this test is presented in Chapter 7. The sensitivity of juvenile striped bass to two individual toxicants, copper and the pyrethroid insecticide esfenvalerate was investigated. The methods used for each test are described below.

Test 1 – Juvenile striped bass, test setup date 7/30/2005. Juvenile striped bass (approximately 3 months old, fork length: 5-5.4 cm) were purchased from Professional Aquaculture Services (Chico, CA), and acclimated to laboratory conditions for 2 days before tests were initiated. Upon arrival, fish were placed into 10-gallon aquaria (30-50 fish per aquarium) containing well water, which had been brought to a salinity of 8 ppt to match the salinity of the water in which the fish were transported. The well water at the UC Davis Center for Aquatic Biology and Aquaculture (UCD CABA) is obtained from a local well approximately 60 m in depth, passed through a packed column aerator to remove excess nitrogen and re-oxygenate, and pumped either directly to the animals or to appropriate cooling and heating equipment. The next day, approximately 80% of the water was replaced with well water salinity-adjusted to 4 ppt, and later in the day with well water diluted with deionized water to a hardness of 200mg/L CaCO₃. This diluted well water was used as control water throughout the experiment.

This 7-day chronic toxicity test measured the effects of Delta water samples on the survival and growth of juvenile *M. saxatilis*. Samples were collected on July 27/28, 2005 from CDFG stations 340, 711, 340 and 915 (see Chapter 3.1), and 7-d tests were initiated on July 30, 2005. Treatments consisted of 5 replicate aquaria, each containing 5 L of aerated water and 5 fish. Water temperature was maintained at 20±1°C. Fish were fed daily (Silver Cup 2.0 mm pellets). The light:dark cycle was 16h:8h. Approximately 80 percent of the water in each replicate was renewed on days 2, 4, and 6 of the test. On days 1, 3, and 5 the numbers of live, dead, and missing fish were recorded. Water temperature, pH, and DO were measured daily. Ammonia nitrogen was measured prior to each water renewal. At test termination, temperature, pH, DO, EC, and ammonia were measured, and one fish per replicate was measured for mass and fork length, and frozen for analysis of tissue chemistry. The remaining 4 fish were measured and individual tissues (brain, kidney, spleen, liver, muscle, gill) dissected, snap-frozen and stored at -80°C for subsequent analyses of sublethal biomarkers.

Test 2 - Larval striped bass test development, test setup date: 6/20/2006. This pilot test was performed using laboratory control water (diluted well water) and 19-d old striped bass larvae obtained from David Ostrach, UC Davis, CA. Striped bass are believed to be highly sensitive to fluorescent light. In order to minimize exposure to fluorescent light, a specialized enclosure was built around an environmental chamber set

to a temperature of 20°C. The tent-like enclosure constructed for this test was assembled with large, black plastic sheets attached to the top and sides of the chamber extending outward to form a large area blocked from light. This allowed the chamber doors to be opened and all necessary test procedures to be performed without leaving the darkened area. To further reduce the risk of exposing the animals to light, the inside of the chamber was lined with black plastic sheets, and illuminated by two night lights.

Fish were transported from adjacent buildings at UCD CABA to UCD ATL in black plastic buckets with closed lids. Tests were performed using four replicate one-liter glass beakers per treatment. Each beaker contained 500ml of well water and ten larvae. Two different loading techniques were tested in an effort to determine the least stressful means of handling the fish, ideally further reducing stress-induced mortality. One treatment was loaded using a modified 5ml glass pipette and the other loaded with an unmodified 5 ml pipette and a pipette pump. The fish were fed approximately 50 artemia twice daily. Water changes (80%) were performed on days two, four and six. Mortality was recorded daily and dead fish were removed. Initial temperature, DO, EC and pH measurements were recorded upon test setup (day 0) and on days 2, 4 and 6. Final temperature, DO and pH measurements (i.e. before exposure water was exchanged) were recorded on days 2, 4 and 6. Final ammonia nitrogen was measured on day 2 and at test takedown.

Test 3 – Larval striped bass, test setup date: 7/14/2006. This test was performed on water samples collected from CDFG stations 340, 508, 609, 711, 910 and 915 on July 11-13, 2006, using 30-d old striped bass larvae obtained from David Ostrach, UC Davis, CA. To avoid exposing the animals to fluorescent light, the entire test was performed in a windowless room, with the lights shut off. Windows to adjacent rooms were covered with black plastic to reduce light entering the room, and entry and exit into the room was restricted. The fluorescent lights were covered with thick black plastic to prevent exposure to light should they accidentally be turned on. Night lights were placed around the 20°C water bath to allow all necessary testing procedures to be performed, while minimizing light-related stress to the animals. Additionally, the exterior door was propped open approximately four inches to allow a small amount of natural light into the room.

Upon receipt, the fish were transferred to four 10-gallon aquaria containing control (diluted well) water. Photos of the gut contents of the larvae were taken under a microscope to monitor feeding before acclimating the animals overnight. Photos were taken daily for the remainder of the test. The following day, all dead fish were removed from the holding tanks. Ten fish were transferred into each of 4 replicate tanks each containing 5 L of control water, then fed 2 ml of artemia/tank and held overnight. Remaining animals were placed in a 10-gallon holding tank for continued monitoring of feeding behavior. At test initiation, 80% of the control water was removed from the treatment tanks and replaced with 5 L of Delta water sample or respective control water. Three controls were included in this test: local well water, a low conductivity control prepared by dilution of well water with glass distilled water to attain a measured conductivity of 100 $\mu\text{S}/\text{cm}$, as well as a high conductivity control of well water, salted up

with Instant Ocean aquarium salt to measure approximately 18.5 mS. Eighty percent water changes were performed on days two, four and six. Mortality was scored daily and dead fish were removed. Initial temperature, DO, EC and pH measurements were recorded upon test setup (day 0) and on days 2, 4 and 6. Final temperature, DO and pH measurements (i.e. before exposure water was exchanged) were recorded on days 2, 4 and 6. Final ammonia nitrogen was measured on day 2 and at test takedown.

Test 4 – Juvenile striped bass, test setup date: July 11-13, 2006. This 7-day chronic toxicity test measured the effects of Delta water samples collected on August 22/23, 2006, from CDFG stations 340, 508, 609, 711, 910 and 915 on the survival and growth of juvenile *M. saxatilis*. Juvenile striped bass (approx. 80 d old, fork length: 5.3 – 8.0 cm) were obtained from David Ostrach, UC Davis. These fish were reared in well water at the UCD CABA facility. Fish were acclimated to laboratory conditions for 1 day before tests were initiated. Upon arrival, fish were placed into 10-gallon aquaria (approximately 30 fish/tank) containing well water for acclimation. Well water was also used as control water throughout the experiment. On the day of test initiation (day 0), tanks filled with 5 L ambient water sample were aerated and brought to the experimental temperature of 20°C. Five fish were then transferred into each of five replicate tanks per treatment. Fish were fed daily with Silver Cup 2.0 mm pellets. The light:dark cycle was 16h:8h. Approx. 80 percent of the water in each replicate was renewed on days 2, 4, and 6. On days 1, 3, and 5, water was not renewed, but the numbers of live, dead, and missing fish were recorded for each replicate. Water temperature, pH, and DO were measured daily. Ammonia nitrogen (NH₃-N) was measured prior to each water renewal. At test termination, temperature, pH, DO, EC, and ammonia were measured for each treatment. On Day 7, fish from each replicate were measured for weight and fork length, and individual tissues (brain, anterior kidney, spleen, liver, muscle, gill) dissected, snap-frozen and stored at –80°C for subsequent analyses of sublethal biomarkers.

Tests 5 and 6 - Exposures to individual toxicants: copper and esfenvalerate. Juvenile striped bass were exposed to copper or the pyrethroid insecticide esfenvalerate [(S)- α -cyano-3-phenoxybenzyl-(S)-2-(4-chlorophenyl)-3-methylbutyrate] in two separate tests. Fish were exposed to CuCl₂*2 H₂O for 7 days, and to esfenvalerate for 24 h, and mortality as well as sublethal endpoints (growth, swimming behavior, transcription of stress response genes) were quantified. The shorter exposure time for the esfenvalerate study is based on the hypothesis that this hydrophobic chemical tends to quickly adsorb to particulate and organic matter in a typical field situation (Yang et al. 2006 a, b; Brady et al. 2006) thus rendering exposure times for fish relatively short.

Juvenile striped bass used in the Cu exposure (90 days old, fork lengths 5.0 – 5.4 cm) were purchased from Professional Aquaculture Services (Chico, CA). Slightly larger, but only 81-day old offspring from the same broodstock (fork lengths 5.3 – 8.0 cm; provided by D. J. Ostrach, UC Davis) were used for the esfenvalerate exposure. Fish used in the copper exposure were slowly acclimated to experimental conditions (conductivity: 890 +/- 20 μ S/cm; hardness: 200 mg/L CaCO₃) over the course of 3 days before tests were initiated. The acclimation and control water was obtained from a local, approx. 60 m deep well, passed through a packed column aerator to remove excess nitrogen and re-oxygenate. Striped bass used in the esfenvalerate exposure were maintained in flow-

through circular tanks containing well water treated as described above for 2 weeks before the tests. Previous fish exposures have shown that stress due to transport and maintenance in the laboratory following the procedures described above is minimal. Fish were loaded into experimental 2.5-gallon aquaria 24 hours prior to testing. Each experimental treatment was comprised of five replicate aquaria containing five animals each. Each tank contained 5L of water at 20°C and was aerated throughout the experiment. Tests were initiated by replacing 80% of the water with experimental copper or esfenvalerate solutions, or control water to yield nominal concentrations of 0 (control), 50, 200, 500 and 1000 µg/L Cu²⁺, or 0 (control), 200 µL/L MeOH (solvent control), 1, 3, 7 and 10 µg/L esfenvalerate. Measured copper concentrations on day 0 were 42, 160, 470, and 900 ppb total Cu²⁺, and 42, 160, 440, and 810 ppb dissolved Cu²⁺. Measured esfenvalerate concentrations on day 0 were 0.64 µg/L, 2.20 µg/L, 4.40 µg/L and 6.50 µg/L.

Experiments were conducted using a light:dark cycle of 16h:8h. During the 7-day copper exposure, fish were fed daily (Silver Cup 2.0 mm pellets). Approximately 80 percent of the water in each replicate was renewed on days two, four and six. On days one, three and five, the numbers of live, dead, and missing fish were scored for each replicate. For the 24 h esfenvalerate exposure, fish were not fed and no water exchange was carried out.

Water temperature, pH, and dissolved oxygen (DO) were measured daily. Ammonia nitrogen (NH₃-N) was measured prior to each water renewal. At test termination, temperature, pH, DO, electric conductivity (EC), and ammonia were measured for each treatment. Overall, no significant deviations between measured water parameters among treatments or replicates were detected.

The number of dead fish was counted at the end of the experiment and surviving fish were sacrificed using an overdose of the anaesthetic MS-222 (Sigma, St Louis, MO, USA) in ice water to minimize degradation of RNA. Fork length (to nearest mm) and weight (to nearest 0.1 g) of each fish were recorded. No significant differences in length or weight were detected between individual treatment groups and controls. During the esfenvalerate exposure, swimming behavior and mortality endpoints were assessed after 4 and 24 h. Swimming behavior was assessed by observing each tank for five minutes. Any pronounced deviation (>1 min) from normal (control) swimming patterns was assessed to be abnormal, e.g. when fish were not able to maintain buoyancy, flipped to their sides, lay on the ground, or repeatedly swam in small circles.

5.1.2 Statistical Analysis

For the test initiated on 7/30/05, modified USEPA standard statistical methods were used to compare the ambient samples to the control (USEPA 2002). These methods were the same as those used to analyze *H. azteca* data in 2006 (see section 4.1.1). The test initiated 6/20/06 examining two alternative methods for transferring the animals was also analyzed using this protocol.

The tests performed in 2006 to examine ambient samples were analyzed using

ANOVA with Tukey's Multiple Comparison procedure to allow comparisons of test organism performance among sample waters, as well as between the controls and the ambient samples. The dilution series examining copper and esfenvalerate toxicity in 2006 were analyzed using USEPA standard protocols, including standard methods of calculating lethal and effective concentrations (USEPA 2002). Statistics for all single-concentration and ambient sample tests were performed using the statistical software JMP v5.0.1. Dilution series data were analyzed with CETIS v1.1.

5.2 Results

Test 1 – Juvenile striped bass, test setup date 7/30/2005. Results of this test are shown in tables C1-1 and C1-2 (Appendix C). There was 100% survival, and no significant effect on body weight and fork length in all treatments. Survival was slightly reduced in water from site 711 (Sacramento River near Rio Vista). Fish exposed to water from site 711 had 96% survival however the effect was not significantly different from controls.

Test 2 - Larval striped bass test development, test setup date: 6/20/2006. Results for our pilot test are shown in tables C3-1 and C3-2 (Appendix C). Survival of striped bass larvae was poor beyond the first 24 h of the test. There was no significant difference between the two transfer methods.

Test 3 – Larval striped bass, test setup date: 7/14/2006. Results for our test on Delta water samples are shown in tables C4-1 and C4-2 (Appendix C). Mean control survival after 96 h was 33% at the low EC (128 $\mu\text{S}/\text{cm}$), 45% at a moderate EC (675 $\mu\text{S}/\text{cm}$) and 75% at the high EC (16,490 $\mu\text{S}/\text{cm}$). Larvae showed highest survival rates in water from site 340 (82%; Napa River). Percent survival in water from this site was significantly higher than percent survival in water from site 915 (28%; Old River-Western arm at railroad bridge) and was likely related to the EC. The EC was 146 $\mu\text{mhos}/\text{cm}$ at site 915, and 15,750 $\mu\text{mhos}/\text{cm}$ at site 340.

Test 4 – Juvenile striped bass, test setup date: 8/25/2006. No significant acute toxicity to juvenile (80-d old) striped bass was observed in samples collected on August 22/23, 2006 from CDFG stations 340, 508, 609, 711, 910 and 915 (Tables C5-1, C5-2, C3-3, Appendix C). Results from the analysis of sublethal biomarker endpoints in striped bass tissues are presented in Chapter 7.

Tests 5 and 6 - Exposures to individual toxicants: copper and esfenvalerate. - Summaries of the effect concentrations of Cu and esfenvalerate on survival of striped bass juveniles are provided in Tables 5-1 and 5-2. Detailed test results are presented in Table C2 (Appendix C). For the Cu exposure, 100% mortality was observed at 470 and 900 $\mu\text{g}/\text{L}$ Cu (440 and 810 $\mu\text{g}/\text{L}$ dissolved Cu), whereas all fish survived in control water and at 42 $\mu\text{g}/\text{L}$ Cu (40 $\mu\text{g}/\text{L}$ dissolved Cu). At a concentration of 160 $\mu\text{g}/\text{L}$ Cu, survival was 92%. The NOEC and LOEC for 96 h and 7 days were the same. No significant effects of Cu exposure on growth or swimming behavior were observed. LC50 and EC25

for total and dissolved Cu in the 7-day exposure were about 60% of those for the 96 hour exposure.

Exposure to esfenvalerate for 24 h resulted in 100% mortality at 4.4 µg/L and 6.5 µg/L esfenvalerate (Table C6, Appendix C). At 2.2 µg/L 24-h survival was 40%. All individuals of control, solvent control and 0.64 µg/L esfenvalerate treatments survived, and only one out of the 25 solvent control fish (4%) showed abnormal swimming behavior. No mortality was observed after 4 hours in any treatment, but abnormal swimming behavior was observed in 76% of striped bass exposed to 6.5 µg/L esfenvalerate, and in 36% of fish exposed to 4.4 µg/L esfenvalerate.

Table 5-1: Effect concentrations of Cu²⁺ on juvenile striped bass (*M. saxatilis*) survival during a 7-day exposure.

Time	Total Cu ²⁺ (ppb)			Dissolved Cu ²⁺ (ppb)		
	LC50	NOEC	LOEC	EC25	NOEC	LOEC
96 hours	441	160	470	414	160	440
7 days	262	160	470	254	160	440

Table 5-2: Effect concentrations of esfenvalerate (µg/L) on juvenile striped bass (*M. saxatilis*) survival during a 24-hour exposure

Time	Survival			Swimming Behavior		
	LC50	NOEC	LOEC	EC25	NOEC	LOEC
4 hours	NA	6.5	> 6.5	3.88	2.2	4.4
24 hours	2.17	0.64	2.2	1.07	0.64	2.2

The contaminants used in this study, copper and esfenvalerate, are known to be toxic to fish, but have very different mechanisms of action. Copper, an abundant heavy metal in the environment (Bielmyer et al., 2006), exerts its toxicity to fish by inhibiting the branchial Na⁺K⁺-ATPase and ion uptake as well as stimulating Na⁺, K⁺ and Cl⁻ efflux from gill surfaces (Lauren and McDonald, 1985). The pyrethroid insecticide esfenvalerate is a potent neurotoxicant that interferes with nerve cell function by interacting with voltage-dependent sodium channels as well as other ion channels, resulting in repetitive firing of neurons and eventually causing paralysis (Bradbury and Coats, 1989). It has been previously shown that juvenile hybrid striped bass (*Morone chrysops* x *Morone saxatilis*) are relatively sensitive to Cu exposure if acclimated to freshwater, with 96-hour acute median lethal concentrations of 94 µg/L (Bielmyer et al., 2006). This concentration is lower than the values observed in this study (414 µg/L), but toxicity of Cu in fish is strongly influenced by environmental parameters such as DOC, pH, hardness and salinity (Reardon and Harrell, 1990; Welsh et al., 1995; Erickson et al.,

1996). Acute toxicity of esfenvalerate in fish occurs at concentrations of approximately 0.1-0.5 $\mu\text{g/L}$ (24-96-h LC50; Siepmann and Holm, 2000; Oros and Werner, 2005). Due to the compound's hydrophobic properties, exposure of aquatic organisms living in the water-column may only be brief (a few hours) or take place via dietary uptake (Werner et al., 2002). The available data suggests that esfenvalerate toxicity to fish is size-dependent. This may explain why the 24-h LC50 of 2.17 $\mu\text{g/L}$ for striped bass juveniles used in this study was higher than reported values.

Sublethal toxic effects can occur at exposure levels far below the concentrations that cause lethality and can have severe consequences for the fitness, reproductive success and survival of aquatic organisms, ultimately leading to population-level effects. For an assessment of the toxic potential of chemicals on fish and aquatic ecosystems, endpoints from laboratory tests such as growth, swimming behavior and molecular stress responses should therefore be interpreted in the context of their environmental consequences. As confirmed in this study, growth endpoints are of limited value for short term (< 7 d) exposures of juvenile striped bass, especially if fish size is not homogenous and if the number of organisms tested must be limited to low numbers. Monitoring of swimming behavior can be a powerful and sensitive biomarker for sublethal effects, as shown for the esfenvalerate exposure. Decreased swimming performance most likely decreases the ability to chase prey or to avoid predation, and is thus an important indicator for overall fitness (Holcombe et al., 1982; Little et al., 1990, Scholz et al., 2000, Sandahl et al., 2005). Non-technical and non-computational methods for the assessment of abnormal swimming behavior, however, are prone to a certain bias depending on the researcher and the time intervals in which they are carried out, and are thus difficult to standardize. Linking results from laboratory exposures to field data is complicated by the fact that it mostly remains untested if fish are able to sense certain chemicals and minimize their exposure by swimming into refuge areas or if they become more vulnerable to predation (Floyd et al., 2008).

5.3 References

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6. Tests with Delta Smelt (*Hypomesus transpacificus*)

Test protocols were developed at UCD-ATL for toxicity tests using delta smelt larvae at different stages of development. While static renewal tests were performed in 2006, a flow-through system was constructed for testing during the 2007 season. This flow-through system proved to be superior to the static renewal method. We included the tests performed in 2005 in this report, because biomarker data for this test is presented in Chapter 7. The sensitivity of delta smelt larvae and juveniles to two individual toxicants, copper and the pyrethroid insecticide esfenvalerate, was investigated. The methods for each test are described below.

6.1 Methods

6.1.1 2005 Toxicity Testing

Juvenile delta smelt 7-day toxicity test: This 7-day chronic toxicity test measured the effects of Delta water samples on the survival and growth of juvenile *H. transpacificus*. Samples were collected on August 30/31, 2005 from CDFG stations 340, 711, 910 and 915, and tests were initiated on September 1, 2005. Each experimental treatment was comprised of 4 replicates of 10 animals each, and each replicate tank contained 7 L of water at 20°C. Fish were fed twice daily with artemia (< 48 hrs old). The light:dark cycle was 16h:8h.

Fish were received 2 days prior to test initiation. Upon arrival, fish were placed into dilute well water in gently aerated test tanks, 10 fish in each 7 liter tank. Reserve fish were placed in a 10 gallon aquarium containing dilute well water (< 100 fish). The day after arrival, 80 percent of the water in each tank was replaced with new dilute well water. This dilute well water was used as control water throughout the experiment.

On the day of test initiation (day 0), 80 percent of the water in each replicate tank was replaced with test water. Fish from the reserve tank were transferred to tanks in which mortality had occurred over the 2 day acclimation period to bring the total number of fish in each replicate to 10. 80 percent of the water in each replicate was renewed on days 2, 4, and 6. On days 1, 3, and 5, water was not renewed, but the numbers of live, dead, and missing fish were scored for each replicate. Water temperature, pH, and DO were measured daily. Ammonia nitrogen (NH₃-N) was measured prior to each water renewal. On Day 7, mass and fork length of 4 fish per replicate were measured and individual tissues (brain, kidney, spleen, liver, gonads, muscle, gill) were dissected, snap-frozen and stored at -80°C for subsequent analyses of sublethal biomarkers. The remaining fish from were measured and frozen whole for chemical analysis. At test termination, temperature, pH, DO, EC, and ammonia were measured for each treatment.

6.1.2 2006 Toxicity Testing

During the 2006 testing season, materials and methods for delta smelt test protocols were refined continuously to incorporate new findings and observations. Control tests were performed to determine the influence of light, water turbidity and conductivity on larval feeding behavior and survival. Methods used are presented below

for each test.

Test organisms: We performed tests using larval delta smelt ranging in age from 9 days old to 92 days old. Delta Smelt were hatched and raised in large tanks at the UC Davis Fish Conservation and Culture Laboratory, Tracy, CA. At this facility, the delta smelt were kept in water pumped directly from the Delta. *Nannochloropsis* algae were added to increase turbidity and *Artemia* were added for food. Younger animals were also fed rotifers.

Control water: Laboratory control water (deionized water amended to US EPA moderately hard specifications, US EPA, 2002) was initially used in control treatments. Since delta smelt larvae did not do well, subsequent tests used water from the delta smelt hatchery for all control treatments. This water is pumped directly from the intake channel of the H.O Banks Pumping Facility near Byron, CA, then passed through a series of sedimentation beds containing natural vegetation to allow any suspended solids in the water to precipitate. This less turbid water is then exposed to an ozonation system to kill any potentially harmful microbes. One day before fish were collected, about 340 gallons of ozonated water were transported to UCD-ATL, and appropriate control waters were prepared for the test.

Fish collection: Fish were maintained in large flow-through tanks at the Byron Hatchery. Using a drain valve, the water was dropped to approximately 1/3 the initial volume of water to increase fish density and thus facilitate collection of the fish. One liter beakers were used to scoop up fish. These were then gently poured into a 27 x 38 cm metal pan containing water at a depth of approximately 2 cm. When the pan contained 30- 40 fish they were then gently poured into black plastic buckets containing hatchery water at a depth of 8-10 cm. Once the desired fish number was reached, the transport bucket was filled to the brim with hatchery water and bucket lids were sealed to prevent water leakage. Dissolved oxygen content was initially monitored during transport. It was not necessary to aerate the water during transport. Buckets were then loaded into coolers packed very lightly with ice to keep temperature at 14-16° C. Small pieces of foam were placed around buckets to reduce vibration. EC and SC were measured in hatchery water. Fish were then transported to the UCD-ATL in Davis. Ice in coolers was replenished periodically during transport to maintain a water temperature of 14-16° C.

Sampling sites: Delta water samples were collected from sites 711, 910, 915, 609, 508 and 340.

Test 1, setup date: 4/5/2006: This seven day test was performed using 9-day old delta smelt larvae. Fish were transported to UCD-ATL in cooled, black 2-gallon buckets with 200 fish per bucket. Upon arrival at the laboratory, fish were placed directly into 2-L test beakers. Larvae were carefully transferred from the black bucket into a glass bread pan using a 250 ml beaker then transferred from there into the test beakers using a 100 ml beaker. Each treatment (six ambient samples, plus control) consisted of four replicate 2-L beakers, each containing 1500 ml of water and ten fish. The fish were fed 1 ml of artemia daily. Tests were performed at 8h:16h D:L cycle, and at a water temperature of 16°C. On

days two, four and six, 80% of the water was exchanged. De-ionized water amended with salts to USEPA moderately hard specifications (DIEPAMH) was used as a control for this test. EC, DO, pH, temperature and ammonia were measured and recorded on days 0, 2, 4, 6 and at test takedown for all treatments.

Test 2 (setup date: 4/18/2006) – Test development: light conditions and turbidity: Poor feeding appeared to have been a problem in the previous tests, potentially resulting in increased mortality. This test was set up to determine optimal light and turbidity conditions for delta smelt larvae. The test was performed using 15-day old delta smelt larvae.

Fish were transported to UCD-ATL in cooled, black 2-gallon buckets with 200 fish per bucket. Upon arrival at the laboratory, fish were placed directly into 2-L test beakers. Larvae were carefully transferred from the black bucket into a glass bread pan using a 250 ml beaker then transferred from there into the test beakers using a 100 ml beaker. Each beaker contained 1500 ml of treatment water and ten fish. Deionized water was amended with salts to USEPA moderately hard specifications (DIEPAMH), and Nanno 3600™, a concentrated *Nannochloropsis* algae solution (68 billion cells per ml; Reed Mariculture, Inc. Campbell, CA) was added to adjust the turbidity of the water (Table 6-1). Each treatment consisted of four replicate beakers. In addition, tests were set up in two different rooms, one with ambient light and one with fluorescent light, to determine the effects of different light conditions on feeding behavior of the smelt larvae. Tests were performed at 8h:16h D:L cycle, and at a water temperature of 16°C. The fish were fed 1ml of artemia and approximately 500 rotifers daily. On days 2, 4 and 6, 80% of the water was exchanged and larvae were checked visually using a flashlight to determine if they were feeding. EC, DO, pH, temperature and ammonia were measured and recorded on days 0, 2, 4, 6 and at test takedown for all treatments.

Table 6-1. Treatment list of delta smelt larvae test #2 to determine optimal lighting conditions and turbidity.

Treatment	Light Conditions	Algae Cells	Turbidity
Hatchery Water Control	Fluorescent Light	0	7.5 NTU
DIEPAMH+1x turbidity ¹	Fluorescent Light	578 x 10 ⁹	7.5 NTU
DIEPAMH+2x turbidity ¹	Fluorescent Light	2.31 x 10 ¹²	15.0 NTU
DIEPAMH+3x turbidity ¹	Fluorescent Light	3.47 x 10 ¹²	22.5 NTU
DIEPAMH Control	Fluorescent Light	0	NA
Hatchery Water Control	Ambient Light	0	7.5 NTU
DIEPAMH +1x algae ¹	Ambient Light	578 x 10 ⁹	7.5 NTU
DIEPAMH +2x algae ¹	Ambient Light	2.31 x 10 ¹²	15.0 NTU
DIEPAMH +3x algae ¹	Ambient Light	3.47 x 10 ¹²	22.5 NTU
DIEPAMH Control	Ambient Light	0	NA

1. De-ionized water amended with salts to EPA moderately hard specifications (DIEPAMH) + algae added to match turbidity, twice the turbidity and three times the turbidity of hatchery control water.

Test 3, setup date: 5/3/2006: This seven day test was performed using 30-day old delta smelt larvae. Fish were transported to UCD-ATL in cooled, black 2-gallon buckets, each containing 250 fish. Upon arrival at the laboratory, fish were placed directly into 2-L test beakers. Larvae were transferred into the test beakers as described above. Each beaker contained 1500 ml of treatment water and ten fish. Each treatment consisted of four replicate beakers. The fish were fed 1 ml and approximately 500 rotifers daily. Tests were performed at 8h:16h D:L cycle, and at a water temperature of 16°C. On days 1, 3 and 5 feeding observations and mortality were recorded. On days 2, 4 and 6, 80% of the water was exchanged. EC, DO, pH, temperature and ammonia were measured and recorded in transport containers upon arrival at the UCD-ATL, and during the test on days 0, 2, 4, 6 and at test takedown for all treatments.

Three controls were used: unmodified hatchery water (“control”), hatchery water taken from the hatchery prior to addition of Nanno 3600™, later modified in the lab to match the lowest turbidity in ambient water samples (“low turbidity control”), and hatchery water diluted to match the lowest conductivity in the ambient samples then modified by addition of concentrated *Nannochloropsis* algae solution to match the turbidity of the hatchery water (“low EC control”). Salinity and turbidity of water samples are shown in table 6-2.

Table 6-2. Salinity and turbidity in in water samples delta smelt larvae test #3.

Site/Treatment	Salinity (ppt)	Turbidity (NTU)
711	0.1	12.7
910	0.1	7.64
915	0.1	6.83
340	0.2	59.2
508	0.1	12.1
609	0.2	29.4
Hatchery Water Control.	0.6	6.01
Low Turbidity Control. ²	0.1	6.00
Low EC Control. ³	0.1	7.86

Test 4, setup date: 5/17/2006: This test was performed on 40-d old delta smelt larvae. Fish were transported to UCD-ATL in black 2-gallon buckets each holding 450 fish. Upon arrival at the laboratory, fish were placed directly into 2-L test beakers using methods described above. Each beaker contained 1500 ml of treatment water and five fish. The smaller number of fish used for this test resulted from high mortality during transport to UCD-ATL, likely due to the high fish density in transport buckets. Each treatment consisted of four replicate beakers. The fish were fed 1 ml and approximately 500 rotifers daily. Tests were performed at 8h:16h D:L cycle, and at a water temperature of 16°C. On days 1, 3 and 5 feeding observations and mortality were recorded. On days 2,

4 and 6, 80% of the water was exchanged. EC, DO, pH, temperature and ammonia were measured and recorded in transport containers upon arrival at the UCD-ATL, and during the test on days 0, 2, 4, 6 and at test takedown for all treatments.

Two controls were used: one control was unmodified hatchery water. The second control was hatchery water diluted to match the lowest EC in ambient samples then modified by addition of concentrated *Nannochloropsis* algae solution to match the turbidity of the hatchery water control. EC was measured in this test instead of salinity for increased accuracy in matching the low EC control. As the hatchery water had the lowest turbidity, the low turbidity control was omitted. Salinity and turbidity of water samples are shown in table 6-3.

Table 6-3. EC and turbidity in water samples of the delta smelt larvae test #4.

Site/Treatment	EC ($\mu\text{S}/\text{cm}$)	Turbidity (NTU)
711	95.6	14.0
910	122.7	8.78
915	122.7	9.73
340	122.7	63.9
508	122.6	13.3
609	354.1	67.5
Hatchery Water Control	668	4.76
Low Conductivity Control	100	4.20

Test 5, setup date: 6/1/2006: This test was performed on 60-d old delta smelt larvae. Fish were transported in a 30 gallon insulated carboy containing 450 fish. A temperature probe was secured in the carboy so that temperature could be monitored during transport. Upon arrival at UCD-ATL, the carboy was placed in the 16°C bath and aerated overnight. The following day, larvae were transferred from the carboy to a large bread pan with a 2L beaker then loaded into 2.5 gallon fish tanks (4 replicates per treatment/3 replicates for low EC control). Treatments consisted of six ambient samples, plus hatchery water control and low EC control. Each tank contained 7 L of treatment water (temperature: 16°C) and ten fish for the duration of the test. The fish were fed 2 ml of artemia twice daily (am and pm). On days 2, 4 and 6, 80% of the water was exchanged. Temperature, EC, DO, ammonia and pH were measured upon arrival at UCD-ATL, as well as on days 0, 2 and 4 of the test. Salinity and turbidity of water samples are shown in table 6-4.

Table 6-4. EC and turbidity in water samples of the delta smelt larvae test #5.

Site/Treatment	EC ($\mu\text{S}/\text{cm}$)	Turbidity (NTU)
711	114.0	15.7
910	131.9	9.2
915	130.2	9.13
340	3596	40.8
508	264.9	12.8
609	623	57.7
Hatchery Water Control	490.5	3.92
Low Conductivity Control	124.3	3.84

Test 6, setup date: 6/15/2006: This 7-d test was performed on 90-d old delta smelt larvae. Fish were transported in a 30 gallon insulated carboy containing 250 fish. A temperature probe was secured in the carboy so that temperature could be monitored during transport. Fish were received 1 day prior to test initiation. Upon arrival, fish were placed into hatchery water (16°C) in gently aerated test tanks, 6-7 fish in each 7 liter tank. The remaining fish were placed in a 10 gallon aquarium containing hatchery water (< 100 fish).

The following day (test day 0), dead fish were removed and 80 % of the water in each replicate tank was replaced with test or control water. Treatments consisted of four replicate 2.5 gallon fish tanks. Fish from the reserve tank were transferred to tanks in which mortality had occurred over the acclimation period to bring the total number of fish in each replicate to 6 (7 for “high EC controls”). Eighty percent of the water in each replicate was renewed on days 2, 4, and 6. The numbers of live, dead, and missing fish were scored daily for each replicate, and dead fish were removed daily. Water temperature, pH, and DO were measured on days 0, 2, 4 and 6. Ammonia nitrogen (NH₃-N) was measured prior to water renewal on day 2 and at test takedown. The fish were fed 2 ml of artemia twice daily. Salinity and turbidity of water samples are shown in table 6-5.

Table 6-5. EC and turbidity in water samples of the delta smelt larvae test #6.

Site/Treatment	EC ($\mu\text{S}/\text{cm}$)	Turbidity (NTU)
711	114.3	3.05
910	135.4	7.83
915	189.8	5.94
340	8320	30.4
508	148.3	9.19
609	202.4	13.76
711 modified to 1500 $\mu\text{S}/\text{cm}$	1474	3.05
Hatchery Water Control	1535	4.79
Low Conductivity Control ³	240.5	4.7
Hatchery Water Control	1535	4.79
DIEPAMH modified to 1500 $\mu\text{S}/\text{cm}$	1493	-

Treatments consisted of six ambient samples collected from various locations within the Delta, plus hatchery water control and low EC control. Two additional controls were tested: 1. The ambient sample with the lowest EC (site 711) was salted up to an EC of 1500 μS using Instant Ocean aquarium salt (“high EC ambient control”, and 2. laboratory control water (DIEPAMH) was salted up to 1500 μS with Instant Ocean aquarium salt. Each additional “high EC” control consisted of 3 replicate aquaria.

6.1.3 2007 Toxicity Testing

During the 2007 testing season, materials and methods for delta smelt flow-through test protocols were developed and refined. Methods used are presented below for each test.

Test organisms and control water: We performed tests using larval delta smelt ranging in age from 21 days old to 92 days old. Delta Smelt were obtained from the UC Davis Fish Conservation and Culture Laboratory, Tracy, CA. Water collected from the hatchery was used for all control treatments (for more detailed description see Chapter 6.1.1).

Fish transport: Fish were transported to UCD-ATL following methods described in Chapter 6.1.1. Test animals collected 4/11/07- 6/22/07 were transported in black 2-gallon (100-150 <45 day old larvae/bucket) placed in coolers packed lightly with ice surrounding the buckets. In later tests (7/26/07 and 8/09/07), 5-gallon buckets (100 54-day old larvae/bucket or 25-40 >54-day old larvae/bucket) were used to accommodate the larger fish.

Sampling sites: For flow-through tests Delta water samples (35 gal per site) were collected from the Hood and Vernalis DWR water quality monitoring stations, and from sites 711, 915, 609, 508 and 340.

Testing procedures: Upon arrival at UCD-ATL, the transport containers with fish were placed into a temperature-regulated water bath maintained at 16° C. One-liter beakers were used to collect fish from the buckets, and fish were gently poured into a metal pan containing water at a depth of approximately 2 cm. The fish were gently scooped up using 100 mL beakers and released into the replicate exposure tanks at random, submerging the beaker and allowing fish to swim freely into the tanks. Twelve fish were placed into each of the test tanks containing 7 L of water for 48-h EC acclimation (Figures 6-1 to 6-3). Sacramento River water, hatchery water and EC-adjusted hatchery water was used as acclimation and control water. EC is adjusted with distilled water (Low EC Control) to match the Sacramento River water samples. When the turbidity of the hatchery water was below 11 NTUs, Nanno 3600™, a concentrated *Nannochloropsis* algae solution (68 billion cells per ml; Reed Mariculture, Inc. Campbell, CA) was added to increase turbidity. Two methods development tests were performed prior to the commencement of ambient testing. Ambient water testing occurred from 5/03/07 to 8/09/07. During acclimation and testing, fish were fed three times a day with 1mL of Artemia and 1mL of rotifers. At test initiation, the EC-adjusted control water is drawn down from 7 liters to approximately two liters to allow for an accurate count of living fish. Water quality parameters (EC, pH, temperature, DO and ammonia concentration) were measured twice daily, and dead fish were counted and removed daily. The feeding behavior of fish was monitored throughout the duration of the test. At test termination, surviving fish were counted.

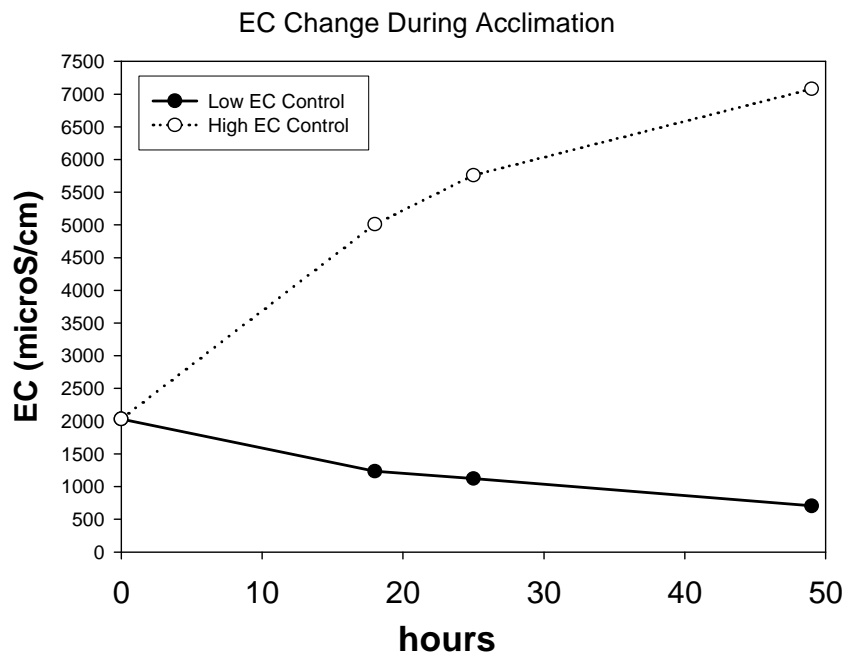


Figure 6-1. EC change in exposure tank during delta smelt 48-h acclimation period to low EC. The flow-through drip system (1.5 ml/min) is used to gradually add EC modified hatchery (control) water to adjust the lowest sample EC.

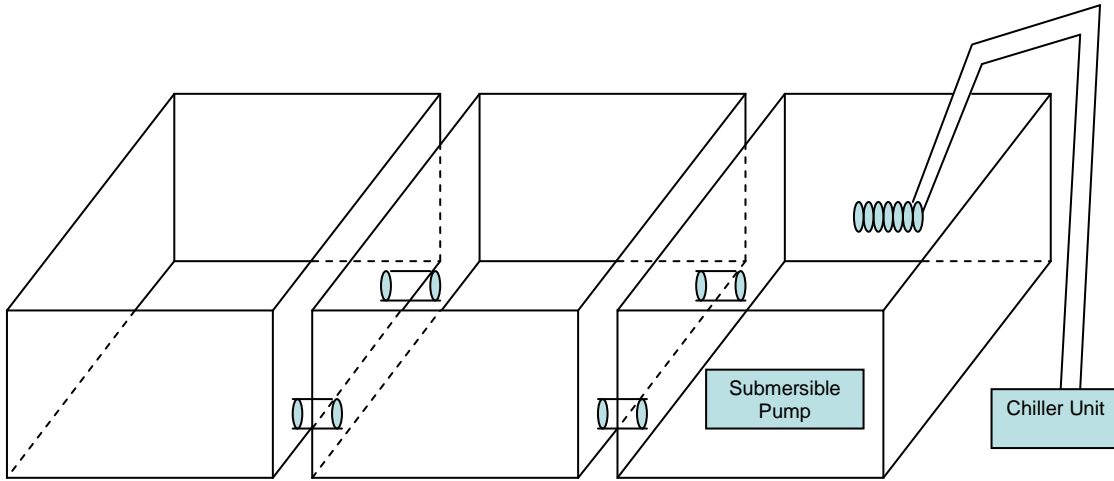


Figure 6-2. The delta smelt flow-through exposure system; diagram shows three 2.5 gallon exposure tanks.

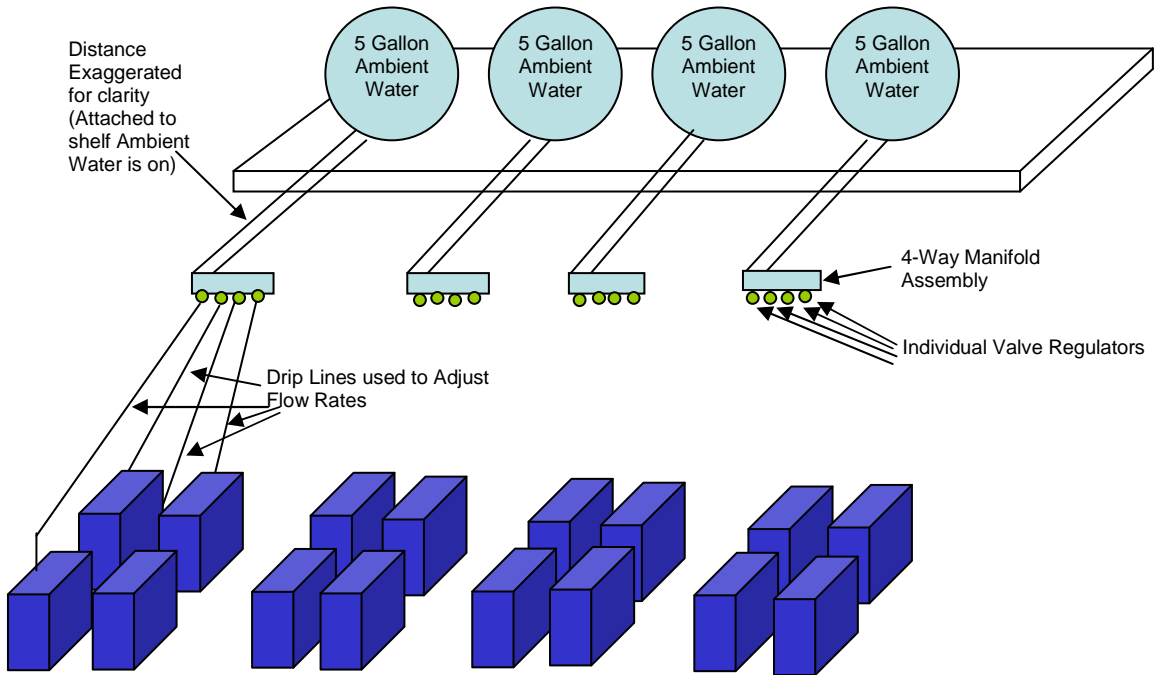


Figure 6-3. Schematic diagram of tank and manifold assembly of delta smelt flow-through exposure system.

Test 1, setup date: 04/26/07: This test was performed using 23-day-old fish collected from the hatchery on 4/26/07. The test was performed in the 16 °C flow-through-system where fish were held in fish breeding nets in 2.5 gallon aquariums containing 7 liters of hatchery water and a small submersible pump. Fish were fed three times a day, with 1mL of Artemia and 1mL of rotifers at each feeding. Upon receipt 10 fish were placed directly into each test apparatus for electrical conductivity acclimation. The flow-through drip system (1.5 ml/min) was used to gradually add EC modified hatchery water in order to alter the electrical conductivity of each treatment to match the electrical conductivity of their respective ambient testing waters for 48 hours, after which the ambient waters would be introduced into the drip system. Periodic electrical conductivity measurements were taken during acclimation. High mortality in the breeding baskets led to the test being terminated during the acclimation phase.

Test 2, setup date: 05/03/07: This test was setup with 28 day old fish collected from the hatchery on 5/01/07. Two replicate tanks were used with 5 fish in each of 2 coffee filter baskets in 2.5 gallon aquariums. Each aquarium contained 7 liters of hatchery water and a small submersible pump in the flow through system. Fish were fed three times a day, with 1mL of Artemia and 1mL of rotifers at each feeding. A second batch of fish was tested in two replicate 600 ml beakers, each containing 250 ml of hatchery water. Fish were fed three times a day, with 100uL of Artemia and 500uL of rotifers at each feeding. After electrical conductivity acclimation, the ambient water samples were introduced to the flow through system via the drip system and to the beakers via an 80% water renewal. In beakers, 80% water changes were performed in every other day. Water chemistry measurements were recorded for all replicates daily.

Test 3, setup date: 05/12/07: This test was set up with 36 day old fish collected from the hatchery on 5/10/07. To minimize handling stress, fish were not loaded directly into the testing tanks, rather were acclimated to the test water ECs in the transport containers modified to fit into the flow through system. Modifications made consisted of a hole in the lid for the drip system and a hole in the side of the buckets for drainage during the EC acclimation period. Fish were fed three times a day, with 5mL of *Artemia* and 5mL of rotifers during acclimation. Three batches of fish were acclimated over a 2 day period to low EC (matching the EC at sites 711 and Hood, 180 uS/cm), medium EC (500 uS/cm), and a high EC (4,700 uS/cm; matching the sites farthest West). One bucket was used to determine if any parts of the flow-through system were inherently toxic to the fish. After two days of acclimation, 10 fish were transferred to 4 replicate 1 liter beakers containing 400 mL of water from Delta sites. For the duration of 7 days, fish were fed three times a day, with 100uL of Artemia and 500uL of rotifers. Mortality was recorded daily. Water renewals (80%) and water quality measurements were performed every other day; ammonia-nitrogen was measured on days 2, 4, 5, 6, and 7.

In addition to the beaker exposures, two experimental procedures were performed. One experimental treatment of tanks tested coffee filter containment units and was not exposed to ambient water samples. These fish were fed three times a day, with 1mL of Artemia and 1mL of rotifers at each feeding. Each tank contained 2 coffee filters, each

containing 7 fish. Two of the replicates (A and B) had a submersible pump to recirculate the water; the other two replicates (C and D) were set up with air-stones for aeration. The second treatment was set up in similar fashion (four replicates exposed to hatchery water in 2.5 gallon tanks) with a second manifold system consisting of airline tubing connected to a simple flow regulator rather than the 4-way manifold with I.V. drip lines used for flow regulation in previous tests. Replicates A and B tested the second manifold with an air-bar with an air-stone in the tank. Fish in these replicates were loose in the tank with no secondary containment. The other two replicates (C and D) had the modified manifold and air-bar assembly, contained 2 coffee filters, each holding 7 fish. At test termination, surviving fish were dried and weighed to determine biomass endpoints.

Test 4, setup date: 05/24/07: This test was set up with 30 day old fish collected from the hatchery on 5/22/07. Upon arrival at the laboratory, 12 fish were immediately placed into the test tanks with no secondary holding units, for EC acclimation. Fish were fed three times a day, with 1mL of Artemia and 1mL of rotifers at each feeding. The changes in electrical conductivity were measured during acclimation. Turbidity was not adjusted for any of the treatments as the hatchery water was higher than 15 NTU. At test initiation, the salinity adjusted control water was drawn down from 7 liters to approximately two liters to allow for an accurate count of living fish. Dead fish were counted and removed daily. At test termination, surviving fish were counted, dried and weighed to determine survival and biomass endpoints.

Test 5, setup date: 06/07/07: This test was set up with 44 day old fish collected from the hatchery on 6/05/07 and brought into the laboratory using transport methods described above. The test used the same methods as the previous test of 052407, with the exception of the addition of *Nannochloropsis* algae and the monitoring of the animals' feeding behavior. *Nannochloropsis* algae concentrate was added to all of the treatments for the entire 2 day acclimation period. After initiation of the ambient water test, *Nannochloropsis* algae were only added to control treatments while ambient sites were completely unaltered. At test initiation the feeding behavior of all the fish was observed and was periodically monitored throughout the duration of the test. At test termination, surviving fish were dried and weighed to determine biomass endpoints.

Test 6, setup date: 06/22/07: This test was set up with 59 day old fish collected from the hatchery on 6/20/07. Testing methods were identical those of the 060707 test. At test termination, the fish were placed into liquid nitrogen and snap frozen by replicate to be stored at -80° Celsius.

Test 7, setup date: 07/26/07: This test was set up with 54 day old fish collected from the hatchery on 7/24/07. Since fish were larger than before, and water quality was a concern, fish were now transported in 4.5-gallon black buckets, as opposed to the 2-gallon buckets used previously. Methods used for ambient sample testing were identical to those in the 5/24/07 test with the exception of the takedown procedures. Turbidity was not adjusted for any of the treatments as the hatchery water was higher than 15 NTU. Due to high mortality in both tests, the tests were taken down early on day 4.

Test 8, setup date: 08/09/07: This test was set up with 92 day old fish collected from the hatchery on 8/07/07. Animals were transported in 5 gallon black buckets. Methods used for this test were identical to those in the 6/07/07 test, with the exception of the termination procedures. After test termination fish were placed into liquid nitrogen and stored at -80°C.

6.1.4 Reference Toxicant Testing

6.1.4.1 Copper

This 7-day chronic toxicity test was conducted to determine the effects of copper on the survival and growth of juvenile *H. transpacificus*. We performed this test two times, once to find the general range of sensitivity of delta smelt to Cu⁺ (rangefinder test), and a second time to determine the LC50 for Cu⁺. Each experimental treatment was comprised of 4 replicates of 10 animals each, and each replicate tank contained 7 L of water at 20°C. The rangefinder test differed from the other test in that each treatment contained only 2 replicates. Fish were fed twice daily with artemia (< 48 hrs old). The light:dark cycle was 16h:8h.

Fish were received 2 days prior to test initiation. Upon arrival, fish were placed into dilute well water in gently aerated test tanks, 10 fish in each 7 liter tank. Reserve fish were placed in a 10 gallon aquarium containing dilute well water (< 100 fish). The day after arrival, 80 percent of the water in each tank was replaced with new dilute well water. This dilute well water was used as control water throughout the experiment.

On the day of test initiation (day 0), 80 percent of the water in each replicate tank was replaced with test water. Fish from the reserve tank were transferred to tanks in which mortality had occurred over the 2 day acclimation period to bring the total number of fish in each replicate to 10. Approximately 80 percent of the water in each replicate was renewed on days 2, 4, and 6. On days 1, 3, and 5, water was not renewed, but the numbers of live, dead, and missing fish were scored for each replicate. Water temperature, pH, and DO were measured daily. Ammonia nitrogen (NH₃-N) was measured prior to each water renewal. On Day 7, 4 fish from each replicate were measured for mass and fork length and individual tissues (brain, kidney, spleen, liver, gonads, muscle, gill) were dissected, snap-frozen and stored at -80°C for subsequent analyses of sublethal biomarkers. The remaining fish from each replicate were measured and frozen for analysis of tissue chemistry. At test termination, temperature, pH, DO, EC, and ammonia were measured for each treatment.

6.1.4.2 Esfenvalerate

This series of experiments was performed to determine the stage-dependent sensitivity of delta smelt larvae to a reference toxicant. The pyrethroid pesticide esfenvalerate was used as a reference toxicant. Delta smelt larvae aged 10 d, 31 d, 35 d, and 52 d were exposed to a range of concentrations for 24 h (see Table 6-6). Larvae were obtained from the UC Davis Fish Conservation and Culture Laboratory, Tracy, CA, and held overnight in the laboratory at 17°C and a 8h:16h D:L light cycle. The following day,

ten larvae were transferred to each 2-L beaker containing 1 L of aerated control water or test solution. Each treatment consisted of 4 replicates, and beakers were shielded with black plastic to provide dim light conditions. Tests were performed at a 8h:16h D:L cycle, and at a water temperature of 16.8-18.2°C. Laboratory control water was prepared according to USEPA protocol with the EC adjusted to hatchery rearing conditions (650 uS/cm – 973 uS/cm @25°C) using Instant Ocean. The pH during the tests was 7.1 – 7.5. Larvae were fed rotifers obtained from the UC Davis Fish Conservation and Culture Laboratory, at 6 pm on the day before initiation of the experiment. They were not fed during the 24-h exposure. Temperature, pH, and dissolved oxygen were measured before and after the exposure in at least 3 randomly selected beakers. Larvae were observed for aberrant swimming behavior, and surviving fish were scored after 4 h and 24 h.

Table 6-6. Esfenvalerate exposure concentrations in reference toxicant tests with different stages of delta smelt larvae.

Delta Smelt Age [d]	Ctr	Solvent Control	0.0312 ug/l	0.0625 ug/l	0.125 ug/l	0.25 ug/l	0.50 ug/l	1.00 ug/l	2.50 ug/l	5.00 ug/l	Temp. °C
10	X	X	X	X	X	X	X				17.6-18.2
31	X	X	X	X	X	X	X	X			16.8-17.5
35	X	X			X	X	X	X	X	X	16.8-17.9
52	X	X	X	X	X	X					17.1-18.2
204	X	X			X	X	X	X	X	X	16.9-17.0

6.1.5 Statistical Analysis

Data from exposures of Delta Smelt were analyzed using both USEPA standard statistical protocols and by one-way ANOVA with Tukey's multiple comparison procedure (USEPA 2002). The USEPA method of data analysis showed the results of the tests according to the standardized statistical method used in aquatic toxicology monitoring and regulation throughout the United States. This method differs from the method used to analyze the 2006 *H. azteca* data, because each comparison of a sample to a control was treated as a separate statistical test, in accordance with USEPA 2002, Appendix H. The Tukey's procedure complemented the USEPA protocol by allowing comparisons other than each treatment paired with one control. Compared to the USEPA procedures, the Tukey's test provided a more conservative evaluation of significant differences between samples since it maintains the experiment-wide alpha at 0.05.

Lethal and sublethal effective concentrations were calculated using CETIS v. 1.1.2 (Tidepool Scientific Software, McKinleyville, CA, USA, 2006). NOEC and LOEC

were calculated using USEPA standard statistical protocols (USEPA 2002). LC50s and EC50s were calculated using linear regression, non-linear regression, or linear interpolation methods. For each endpoint, toxicity is defined as a statistically significant difference ($p < 0.05$) to the laboratory control. Methods used in the analysis of long-term patterns and trends included pairwise correlations, ANOVA, ANCOVA, MANOVA, linear regression, and polynomial regression models performed in JMP 5.0.1.

6.2 Results

6.2.1 2005 Toxicity Testing

Delta Smelt Juveniles, 7-day Test: Results are presented in Appendix D, Tables D3-1 and D3-2. Fish in control water had 95% +/- 2.9% survival, and survival was equal or better at sites 711, 910 and 915 (Table 6-7). Survival of fish exposed to water from site 340 was slightly reduced to 85% +/- 2.9%, but the difference to control was not statistically significant. Fork length and wet weight were similar in all treatments. EC of the water from site 340 was 19 mS/cm, which corresponds to a salinity of approximately 11 ppt.

6.2.2 2006 Toxicity Testing

Results of ambient water tests with delta smelt performed in 2006 are presented in Tables D4-D8 (Appendix D). Survival in water collected May 1, 2006 from site 508 was significantly lower than in other treatments with similar EC (Table D5, Appendix D). Otherwise, no field site-specific toxicity was detected in these tests.

The main water quality parameters correlated to decreased survival of delta smelt larvae were un-ionized ammonia with an estimated effect concentration of >0.012 mg/L NH_3 , Figure 6-4) and electrical conductivity (EC, Table 6-7). Fish survival tended to be highest in water from site 340 (Napa River), which was the site with highest EC. Fish age was a significant factor in survival to day 6 and day 7 under laboratory test conditions, indicating that older larvae were less sensitive to stress due to transport, handling and test conditions than younger larvae. Further analyses and results on the effects of ammonia are presented and discussed in more detail in Chapter 6.2.4. Overall, it was determined that the static renewal testing protocol in 2-liter beakers did not yield satisfactory survival of delta smelt larvae, and a flow-through system was subsequently constructed and used in 2007.

Table 6-7. Pairwise correlations of delta smelt survival with fish age and water quality parameters for the entire 2006 data set. Significant correlations are indicated in bold.

		Maximum Un-ionized NH ₃	Initial EC	Turbidity	Fish Age
Survival	Day 2	0.2834	0.1771	-0.0132	-0.3516
	Day 4	-0.6464	0.2912	0.3431	-0.0009
	Day 6	-0.5517	0.4195	0.2629	0.5103
	Day 7	-0.4261	0.4541	-0.1036	0.5566

6.2.3 2007 Toxicity Testing

Detailed results of toxicity tests with delta smelt larvae performed in 2007 are presented in Appendix D, Tables D9-D14. Results are summarized in Table 6-8. Turbidity and EC/salinity were the two most important factors determining survival of delta smelt larvae overall, particularly for larvae less than 44 days old. These younger larvae generally survived poorly in low EC samples from the lower Sacramento River, Old River and the San Joaquin River, as well as in the low EC control (150-180 uS/cm) even when turbidity was adjusted to 10 NTU. Larvae that were 30-36 d old survived best in water from the Napa River (site 340), and Montezuma Slough (site 609), which had both saline (EC>4000 uS/cm) and the most turbid water. Larvae that were 44 d old and older appeared to be less dependent on high turbidity and salinity, but survival in water samples from the lower Sacramento River was generally lower than in controls or samples from other Delta sites.

Survival was significantly lower among smelt exposed to water samples from Hood collected June 6, 2007 (Table D11-1, Appendix D) and site 711 collected July 26, 2007 (Table D13-1, Appendix D) than in low EC control water. Both sites are located on the lower Sacramento River. Samples collected at Hood and Site 711 on 6/6/07 had very similar EC and unionized ammonia concentrations (Tables D11-2, -3, Appendix D). Turbidity was higher at Hood than at site 711, and similar to site 915 where survival was 87%. It is therefore likely that other factors were responsible for the reduced survival in water from the Hood site. The sample taken at site 711 on July 26 had the same turbidity as Hood and site 915 (Tables D13-2, -3, Appendix D), and the same EC as the low EC control and Hood, thus neither EC nor turbidity appear to be responsible for the low survival. Further analyses and results on the effects of ammonia are presented and discussed in more detail in Chapter 6.2.4.

Table 6-8. Percent survival of delta smelt larvae after 7-day exposures in Delta water samples (unless indicated otherwise). Results indicated in bold/shaded box are significantly different from their respective control. Sites 711 and 915, Hood and Vernalis were compared to the Low EC Control, sites 609, 508 and 340 were compared to the High EC Control.

Sampling Date:	May	May	Jun	Jun	Jul	Aug
	8-10	23-24	5-6	20-21	25-26	8-9
Age of DS Larvae:	36 d	30 d	44 d	59 d	54 d	92 d
Treatment	Mean 7-day Survival (%)					
					[96 h]	
Low EC Control	21	32	89	85	61	82
Site 711	20	37	75	73	29¹	59
Hood	45	41	68	80	57	63
Site 915	45	32	87	75	52	75
Vernalis	45	39	87	-	66	97
Hatchery Control	39	53	98	82	41	92
High EC Control	73	51	94	94	27 ²	94
Site 609	51	90	94	89	41	91
Site 508	43	70	94	92	33	93
Site 340	89	87	89	95	58	82
Low Turbidity Ctr (1.1-1.5 NTU)				83	25 ²	71

¹ Turbidity of site 711 sample was same as Hood sample (3.1 NTU)

² Samples had lowest turbidity 1.3-1.6 NTU

6.2.4 Ammonia and Delta Smelt Survival

6.2.4.1 Testing Period 2006

Figure 6-4 shows the results of a regression analysis on 96-h survival of delta smelt larvae at different ages, and the maximum measured unionized ammonia (mg/L) during the static renewal tests (ambient samples and controls) performed in 2006. In order to measure the response of the healthiest animals possible, this analysis includes only data from experiments showing at least 50% survival in controls on day 7 of the test. Each point represents the mean 96-h survival (n=4 per treatment) of one treatment (i.e. control or ambient sample). Each symbol type corresponds to a different experiment. The experiments were initiated between 4/18/06 – 6/15/06. Results show a significant linear relationship between 4-day survival and unionized ammonia concentration with an estimated 96-h LC50 of 0.012 mg/L NH₃. However, it is important to remember that the laboratory tests were carried out with delta smelt larvae of different ages (9-90 d old), and additional tests to determine ammonia toxicity for delta smelt larvae of single age groups are advisable.

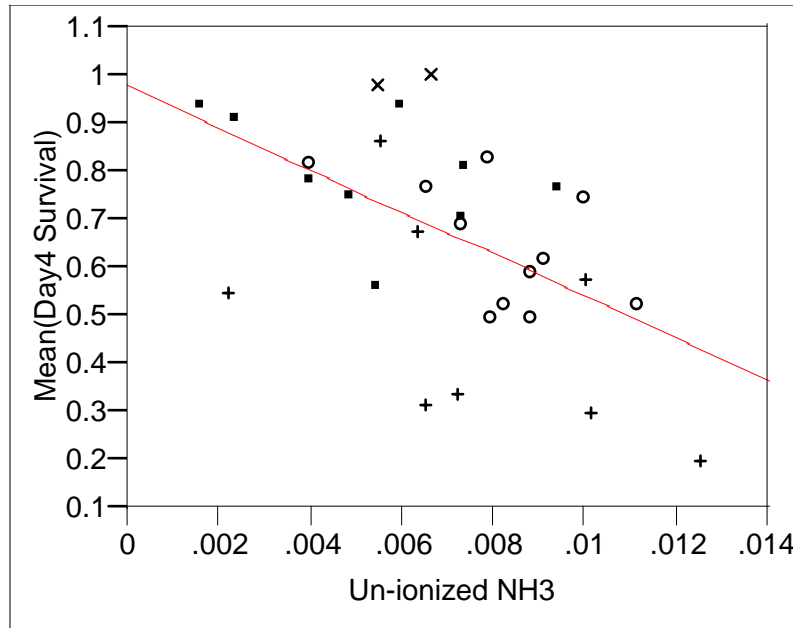


Figure 6-4. Linear fit of mean 4-day delta smelt survival and maximum unionized NH3 measured in test beakers. Mean (4-d Survival) = 0.9787573 - 43.849843 NH3; p<0.002.

Summary of Fit

RSquare	0.296599
RSquare Adj	0.271478
Root Mean Square Error	0.182014
Mean of Response	0.671333
Observations (or Sum Wgts)	30

Analysis of Variance

Source	DF	Sum of Squares	Mean Square	F Ratio
Model	1	0.3911435	0.391143	11.8066
Error	28	0.9276171	0.033129	Prob > F
C. Total	29	1.3187606		0.0019

Parameter Estimates

Term	Estimate	Std Error	t Ratio	Prob> t
Intercept	0.9787573	0.095442	10.26	<.0001
Un-ionized NH3	-43.84984	12.76161	-3.44	0.0019

6.2.4.2 Testing Period 2007

Although toxicity to delta smelt larvae was primarily observed at sites where ammonia concentrations were consistently among the highest (sites 711, Hood), data analysis of delta smelt 7-d survival and unionized ammonia concentrations (maximum

laboratory value measured during 7-day test) showed no correlation (Figure 6-5). However, it is important to remember that the laboratory tests were carried out with delta smelt larvae of different ages (30-92 days old), and further tests to determine ammonia toxicity for delta smelt are advisable.

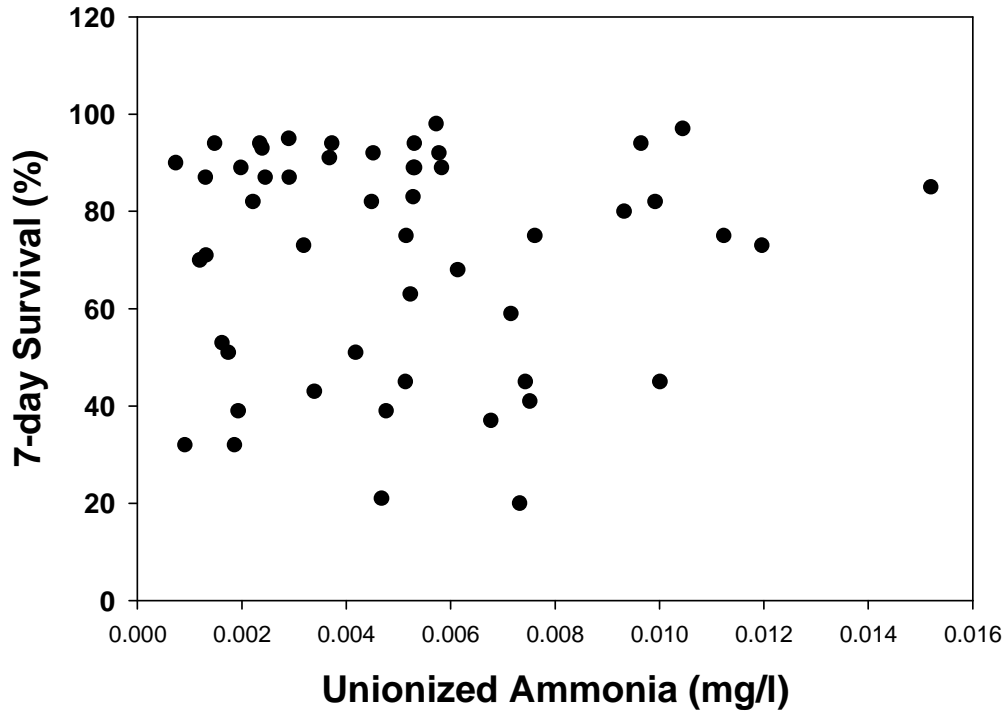


Figure 6-5. No correlation was found between 7-d survival of delta smelt larvae and maximum unionized ammonia concentration in tests performed in 2007.

6.2.4.3 Testing Period 2006-2007

Although we found no correlation between ammonia concentrations and delta smelt survival in 2007, the 2006 data indicated the delta smelt larvae could potentially be highly sensitive to unionized ammonia. In addition, it was noted that the field sites with highest ammonia concentrations had the highest incidence of toxic events, thus the possible effect of ammonia on survival of delta smelt was examined in more detail.

Table 6-9 shows the results of an ANOVA analysis of the complete 2006-2007 data, by year and for both years combined. Unionized ammonia was not found to be positively or negatively correlated with 7-d survival in any dataset. Pairwise correlations showed that EC had a dramatic effect on the survival of the smelt. However, no correlation was found between smelt survival and the turbidity of the water in the exposure tanks. Variable survival due to the effects of EC differences between samples,

the response of the larvae to turbidity, and the inherently variable age/robustness of delta smelt in different experiments might have obscured any effects of ammonia. EC was positively associated with 7-day survival, and 7-day survival varied significantly among batches of smelt (EC effect: linear regression, $F_{1,70} = 39.21$, $P < 0.0001$; Batch effect: one-way ANOVA, $F_{7,64} = 12.77$, $P < 0.0001$). These findings led us to include both mean EC and “experiment membership” as factors in ANOVA models testing for possible effects of ammonia on 96-h and 7-day survival. Experiment membership was included as a categorical covariate, while log-transformed EC was included as a continuous covariate.

The ANOVA models indicated that once conductivity and experiment membership are taken into account, and for the complete 2006-2007 dataset, ammonia did not have a significant effect on delta smelt survival in our tests, where maximum unionized ammonia concentrations were <0.016 mg/L. However, in 2006 we continued to see a marginally significant ($p=0.06$) correlation of 7-d survival and unionized ammonia.

Table 6-9. Direction and strength of associations between delta smelt survival and ammonia concentrations (ammonia-N and NH₃); pairwise correlations and effects in the ANOVA models

Year	Variable	by Variable	ANOVA	
			Effect Coefficient	P
2006 2007	- Smelt 7-d Surv	Log Mean NHN	10.0	0.147
	Smelt 7-d Surv	Log Max NHN	10.1	0.184
	Smelt 7-d Surv	Log Mean NH ₃	1.5	0.646
	Smelt 7-d Surv	Log Max NH ₃	0.8	0.909
2006	Smelt 7-d Surv	Log Mean NHN	75.8	0.152
	Smelt 7-d Surv	Log Max NHN	-46.9	0.317
	Smelt 7-d Surv	Log Mean NH ₃	-11.1	0.129
	Smelt 7-d Surv	Log Max NH ₃	-30.4	0.061
2007	Smelt 7-d Surv	Log Mean NHN	7.1	0.313
	Smelt 7-d Surv	Log Max NHN	10.3	0.185
	Smelt 7-d Surv	Log Mean NH ₃	4.4	0.242
	Smelt 7-d Surv	Log Max NH ₃	7.1	0.366

6.2.5 Reference Toxicant Testing

6.2.5.1 Copper

Delta smelt juveniles were highly sensitive to copper (Tables D15, D16, Appendix D). Data analysis yielded LC50 values for copper ion toxicity of 33.5 µg/L (96 h) and 24.7 µg/L (7 d).

6.2.5.2 Esfenvalerate

A series of 24-h laboratory tests demonstrated that sensitivity of delta smelt larvae to the pyrethroid insecticide, esfenvalerate, was age-dependent (Figure 6-6, Table 6-10). Detailed test results are presented in Tables D17-D20 (Appendix D). The high sensitivity of 52-d old larvae may reflect the fact that at this stage, when swim-bladder inflation occurs, fish may be more sensitive overall to stressful conditions.

The 24-h LC50 for 10-d to 204-d old delta smelt ranged from 0.1-0.76 µg/L esfenvalerate (nominal concentration), and the 24-h EC25 for swimming impairment ranged from 0.03 to 0.28 µg/L esfenvalerate (Table 6-10). The lowest effect concentrations (LOEC) for swimming ability after 24 h ranged from 0.0625 - 0.25 µg/L (Tables 6-11 to 6-15). This indicates that delta smelt larvae are highly sensitive to this pyrethroid insecticide, and due to impairment of their swimming performance may be more susceptible to predation at concentrations as low as 62.5 ng/L esfenvalerate (Floyd et al., in press). However, toxicity of pyrethroids in the Delta is likely alleviated by the presence of particles and organic matter, and to date concentrations of pyrethroids detected in the water column were below this effect concentration.

Table 6-10. Summary of effect concentrations of esfenvalerate for delta smelt larvae and juveniles.

Test	Fish Age	Survival LC50 (ug/L)		Swimming EC25 (ug/L)	
		24 hr	4 hr	4 hr	24 hr
5/17/2006	35 days	0.1	-	-	0.03
5/23/2006	10 days	0.19	0.38	0.38	0.04
5/23/2006	52 days	0.24	0.13	0.13	0.11
6/1/2006	31 days	0.54	1.05	1.05	0.12
10/18/2006	204 days	0.76	1.46	1.46	0.28

Table 6-11. Results of 24-h exposure to esfenvalerate using 10-d old delta smelt. Nominal esfenvalerate concentrations were 0.03125 ug/l – 0.5 ug/l. Endpoints quantified were swimming and survival after 4 and 24 h.

Endpoint	Result of Statistical Analysis
Control – solvent control	Non-sign. 4 hour swimming Non-sign. 4 hour survival Non-sign. 24 hour swimming Non-sign. 24 hour survival
4 hour survival	
4 hour swimming	Significant effects at 0.25 and 0.5
24 hour survival	Significant effects at 0.125 and 0.25
24 hour swimming	Significant effect at 0.0625 and 0.125
NOEL 4 hrs survival	0.5
LOEL 4 hrs survival	>0.5
NOEL 4 hrs swimming	0.125
LOEL 4 hrs swimming	0.25
NOEL 24 hrs survival	0.625
LOEL 24 hrs survival	0.125
NOEL 24 hrs swimming	0.03125
LOEL 24 hrs swimming	0.0625

Table 6-12. Results of 24-h exposure to esfenvalerate using 31-d old delta smelt. Nominal esfenvalerate concentrations were 0.03125 ug/l – 1 ug/l. Endpoints quantified were swimming and survival after 4 and 24 h.

Endpoint	Result of Statistical Analysis
Control – solvent control	Non-sign. 4 hour swimming Non-sign. 4 hour survival Non-sign. 24 hour swimming Non-sign. 24 hour survival
4 hour survival	Non-significant effects between solvent control and 0.03125, 0.0625, 0.125, 0.25, 0.5, 1.0 ug/l
4 hour swimming	Significant effects at concentrations 0.5 ug/l and 1 ug/l
24 hour survival	Significant effects at 0.5 and 1.0 ug/l
24 hour swimming	Significant effects at 0.125 and 0.25 ug/l, higher conc. died and non-testable
NOEL 4 hrs survival	1
LOEL 4 hrs survival	>1
NOEL 4 hrs swimming	0.25
LOEL 4 hrs swimming	0.5
NOEL 24 hrs survival	0.25
LOEL 24 hrs survival	0.5
NOEL 24 hrs swimming	0.0625
LOEL 24 hrs swimming	0.125

Table 6-13. Results of 24-h exposure to esfenvalerate using 35-d old delta smelt. Nominal esfenvalerate concentrations were 0.125 ug/l – 5 ug/l. Endpoints quantified were swimming and survival after 24 h.

Endpoint	Result of Statistical Analysis
Control – solvent control	Non significant for 24 swimming and survival
24 hour survival	Significant effects at 0.125, 0.5, 1.0 ug/l, higher conc not testable (full mortality)
24 hour swimming	Significant effect at 0.125, higher conc not testable
NOEL 24 hrs survival	<0.125
LOEL 24 hrs survival	0.125
NOEL 24 hrs swimming	<0.125
LOEL 24 hrs swimming	0.125

Table 6-14. Results of 24-h exposure to esfenvalerate using 52-d old delta smelt. Nominal esfenvalerate concentrations were 0.03125 ug/l – 0.25 ug/l. Endpoints quantified were swimming and survival after 4 and 24 h.

Endpoint	Result of Statistical Analysis
Control – solvent control	Non-sign. 4 hour swimming Non-sign. 4 hour survival Non-sign. 24 hour swimming Non-sign. 24 hour survival
4 hour survival	Non-signif
4 hour swimming	Significant at 0.25
24 hour survival	Signif. At 0.25
24 hour swimming	Non-signif. At 0.03125, 0.0625, 0.125
NOEL 4 hrs survival	0.25
LOEL 4 hrs survival	>0.25
NOEL 4 hrs swimming	0.125
LOEL 4 hrs swimming	0.25
NOEL 24 hrs survival	0.125
LOEL 24 hrs survival	0.25
NOEL 24 hrs swimming	0.125
LOEL 24 hrs swimming	0.25

Table 6-15. Results of 24-h exposure to esfenvalerate using 204-d old delta smelt. Nominal esfenvalerate concentrations were 0.1 ug/l – 5.0 ug/l. Endpoints quantified were swimming and survival after 4 and 24 h.

Endpoint	Result of Statistical Analysis
Control – solvent control	Non-sign. 4 hour swimming Non-sign. 4 hour survival Non-sign. 24 hour swimming Non-sign. 24 hour survival Non-sign. Length Non-sign. Weight
4 hour survival	Non-significant effects between solvent control and all concentrations.
4 hour swimming	Significant effects at concentration 5.0 ug/l.
24 hour survival	Significant effects at 1.0 ug/l and 5.0 ug/l.
24 hour swimming	Significant effects at 0.25 ug/l, 0.5 ug/l, 1.0 ug/l and 5.0 ug/l.
24 hour length	Non-significant effects between solvent control and all concentrations.
24 hour weight	Non-significant effects between solvent control and all concentrations.
NOEL 4 hrs survival	5
LOEL 4 hrs survival	>5
NOEL 4 hrs swimming	1.0
LOEL 4 hrs swimming	5.0
NOEL 24 hrs survival	0.5
LOEL 24 hrs survival	1.0
NOEL 24 hrs swimming	0.1
LOEL 24 hrs swimming	0.25

6.3 References

- Floyd E.Y., Geist J.P., Werner I. (in press). Short-term exposure of the fathead minnow (*Pimephales promelas*) to a pyrethroid insecticide: Implications for growth, behavior, and predation risk. *Environmental Toxicology and Chemistry*.
- US EPA, 2002. Short-term Methods for Estimating the Chronic Toxicity of Effluents and Receiving Waters to Freshwater Organisms, 4th Ed. USEPA Office of Water, Washington, DC. EPA-821-R-02-013.

7. Sublethal Indicators of Contaminant Effects in Delta Species

7.1 Inhibition of Acetyl-Cholinesterase in Brain and Muscle Tissue of Juvenile Striped Bass and Delta Smelt Exposed to Delta Water Samples and Copper

For organophosphate (OP) and carbamate insecticides, the primary mechanism of toxic action is the inhibition of the enzyme acetylcholinesterase (AChE), which is commonly used as a diagnostic tool for sublethal OP and carbamate exposure and effect. Studies in fish have shown that brain AChE inhibition in excess of 70% is strongly correlated with imminent mortality (Fulton and Key, 2001) however fish are far less sensitive to these groups of insecticides than invertebrates such as crustaceans and insects. For example, Wheelock et al. (2005) report that exposure to 7.3 $\mu\text{g/L}$ CP, a concentration that caused 20% mortality in juvenile Chinook salmon, severely inhibited AChE activity in brain (by 85%) and muscle (by 92%). While all fish survived an exposure to 1.2 $\mu\text{g/L}$ CP, AChE activity in the brain was reduced by 8%. Monitoring studies performed in the 1990s linked toxicity to aquatic life in the Sacramento-San Joaquin Delta and the San Joaquin River basin to OP insecticides (Werner et al., 2000; Domagalski et al., 2000; Dubrovsky et al., 1998), and CP was among the most commonly detected toxicants (Werner et al., 2000) with concentrations of ≤ 0.52 $\mu\text{g/L}$ in the Delta. Elsewhere, concentrations of up to 3.2 $\mu\text{g/L}$ CP have been reported (Salinas River, CA; Hunt et al., 2003). For this study, we quantified AChE activity in brain and muscle of juvenile striped bass and delta smelt exposed to water samples from the Delta or to different concentrations of copper.

7.1.1 Methods

Juvenile delta smelt and striped bass were exposed to Delta water samples, as well as a range of copper concentrations in 2005 (see Chapters 5 and 6), and tissues were dissected, flash-frozen and stored at -80°C . Fish brains were removed entirely, whereas muscle samples consisted of one piece of epaxial white muscle taken from behind the head. Each sample was weighed, diluted 1:10 (mg:l) in 0.1M sodium phosphate buffer (pH 8.0) with 0.5% Triton X-100. Tissues were homogenized for 1 min using a glass douncer on ice. Homogenates were centrifuged at 4°C for 10 min at $7000\times g$ to remove large particulate material. The supernatant fraction was transferred to a separate tube and the total protein concentration was determined with the Biorad DC Protein Assay (Bio-Rad Laboratories, Hercules, CA) using methods of Lowry et al. (1951). For the AChE assay, 0.1M sodium phosphate buffer (pH 8.0) with 0.5% Triton X-100 was added to the supernatant fractions to produce final dilutions of 1:500 (mg:L) for muscle samples and 1:200 (mg:l) for brain samples. Assay optimization was performed with brain and muscle tissue from unexposed juvenile fish. Acetylthiocholine iodide (AtChI) concentrations between 0.1 and 5mM were tested for optimal substrate concentration, and samples were incubated with tetraisopropylpyrophosphoramidate (iso-OMPA, a selective AChE inhibitor) to measure butyrylcholinesterase-mediated substrate hydrolysis. Results showed negligible butyrylcholinesterase activity in muscle tissue, therefore subsequent assays were performed without the AChE inhibitor. AChE activity in brain and muscle was analyzed using modified methods of Ellman et al. (1961). AChE activity for each sample was determined by adding 30l of diluted supernatant to a microplate well (Costar 96 well EIA/RIA Plate; Corning Inc., New York, NY)

containing 250 l of 0.1M sodium phosphate buffer (pH 8.0), 10µl of 5,5-dithiobis-2-nitrobenzoic acid (DTNB, 10.3 mM), and 30 µL of AtChI (21.4 mM). Final assay concentrations were 0.32 mM DTNB and 2mM AtChI. Final protein concentrations ranged from 10.8 to 17.1µg/L for muscle and 7.0 to 10.7µg/L for brain. All assays were performed in triplicate. Absorbance at 412 nm was measured at 2 min intervals for 10 min at 25 °C with an automated microplate reader (Model EL3401; Bio-Tek Instruments, Winooski, VT) and all samples were corrected for background hydrolysis. AChE activity was calculated as mol/min/g wet weight, and then normalized to the amount of protein in the homogenate (mol/min/mg protein).

7.1.2 Results

7.1.2.1 Striped Bass

Ambient Samples: Exposure (7 d) of juvenile striped bass to water samples collected from sites 915 (Old River), 711 (Sacramento River at Rio Vista), 609 (Montezuma Slough) and 340 (Napa River) on July 27/28, 2005, did not affect AChE activity in brain tissue of striped bass (Table 7-1). These fish also showed 100% survival and no significant effect on body weight and fork length in all treatments.

Table 7-1. AChE activity in brain tissue of juvenile striped bass (3 months old) exposed to water samples collected on July 27/28, 2005 at CDFG stations 340, 711, 609 and 915. SD=standard deviation of the mean.

Treatment	Mean Activity (µmol/min/g wet weight)	SD	n
0-Time Control	0.168	0.065	5
Control	0.199	0.022	10
Site 915	0.172	0.060	10
Site 711	0.170	0.086	10
Site 609	0.202	0.081	10
Site 340	0.195	0.045	10

Copper: The highest copper concentration (200 ppb) where 100% of the fish survived the exposure did not have an effect on AChE activity in brain tissue of exposed fish (Table 7-2). The LC50 values determined for Cu⁺ were 348 µg/L (96 h) and 301 µg/L (7 d).

Table 7-2. AChE activity in brain tissue of juvenile striped bass (3 months old) exposed to different copper concentrations for 7 days. SD=standard deviation of the mean.

Treatment	Mean Activity (umol/min/g wet weight)	SD	n
Control	0.350	0.048	10
200 ppb Cu ⁺	0.374	0.053	10

7.1.2.2 Delta Smelt

Ambient Samples: Water samples collected from sites 915 (Old River), 711 (Sacramento River at Rio Vista), 609 (Montezuma Slough) and 340 (Napa River) on August 30/31, 2005, did not affect AChE activity in brain tissue of delta smelt juveniles (Table 7-3). Fish in control water had 95% +/- 2.9% survival, and survival was equal or better at sites 711, 910 and 915. Survival of fish exposed to water from site 340 was slightly reduced to 85% +/- 2.9%, but the difference to control was not statistically significant. Fork length and wet weight were similar in all treatments.

Table 7-3. AChE activity in brain tissue of juvenile delta smelt (90-days old) exposed to water samples collected on August 30/31, 2005 from CDFG stations 340, 711, 910 and 915. SD=standard deviation of the mean.

Treatment	Mean Activity		n
	(umol/min/g wet weight)	SD	
Control	0.227	0.063	8
Site 915	0.386	0.110	8
Site 711	0.291	0.093	8
Site 609	0.310	0.029	8
Site 340	0.276	0.144	7

Copper: Copper did not affect AChE activity at sublethal Cu⁺ concentrations, however, the 50 ppb Cu⁺, which was above the LC50 determined for juvenile delta smelt, significantly reduced enzyme activity in the brain (Table 7-4) but not in muscle tissue (Table 7-5).

Table 7-4. AChE activity in brain tissue of juvenile delta smelt (3 months old) exposed to different copper concentrations for 7 days. The LC50 values for copper ion toxicity were 33.5 µg/L (96 h) and 24.7 µg/L (7 d). SD=standard deviation of the mean.

Treatment	Mean Activity		n
	(umol/min/g wet weight)	SD	
0-Time Control	0.228	0.092	5
Control	0.403	0.131	8
10 ppb Cu	0.388	0.124	8
25 ppb Cu	0.305	0.164	8
50 ppb Cu	0.093	0.103	5

Table 7-5. AChE activity in muscle tissue of juvenile delta smelt (3 months old) exposed to different copper concentrations for 7 days. The LC50 values for copper ion toxicity were 33.5 µg/L (96 h) and 24.7 µg/L (7 d). SD=standard deviation of the mean.

Treatment	Mean Activity (umol/min/g wet weight)	SD	n
0-Time Control	0.333	0.127	5
Control	0.479	0.162	8
10 ppb Cu	0.532	0.172	10
25 ppb Cu	0.479	0.110	10
50 ppb Cu	0.452	0.126	5

7.2 Expression of Stress response Genes in Striped Bass

7.2.1 Comparisons of tissue-specific transcription of stress response genes with whole animal endpoints of adverse effect in striped bass (*Morone saxatilis*) following treatment with copper and esfenvalerate.

Juergen Geist, Inge Werner, Kai J. Eder, Christian M. Leutenegger (2007); published in *Aquatic Toxicology* 85:28–39.

See Appendix G.

7.2.2 Tissue-Specific Expression of Stress response Genes in Striped Bass Exposed to Water Samples from the Sacramento-San Joaquin Delta

7.2.2.1 Methods

Fish exposures: This 7-day chronic toxicity test measured the effects of Delta water samples collected on August 22/23, 2006, from CDFG stations 340, 508, 609, 711, 910 and 915 on the survival and growth of juvenile *M. saxatilis*. Juvenile striped bass (approx. 80 d old, fork length: 5.3 – 8.0 cm) were obtained from David Ostrach, UC Davis. These fish were reared in well water at the UCD CABA facility. Well water was also used as acclimation and control water in the experiment. Laboratory water conditions were adjusted to match the conductivity (890 +/- 20 µS/cm) in which the striped bass were maintained and fish were additionally acclimated to experimental 10-gal aquaria (30 fish/aquarium) for 24 h before tests were initiated and then loaded into experimental tanks 24 hours prior to testing. Each experimental treatment was comprised of five replicate tanks containing five animals each. Each tank contained 5L of water at 20° C and was aerated throughout the experiment. Previous experience in fish exposures

has shown that stress due to fish transport, maintenance of fish in the laboratory and practicability criteria for carrying out toxicological tests are matched well by this procedure. Tests were initiated the next day by replacing 80% of the water with ambient water samples. Experiments were conducted using a light:dark cycle of 16h:8h. During the 7-day copper exposure, fish were fed daily (Silver Cup 2.0 mm pellets). Approximately 80 percent of the water in each replicate was renewed on days two, four and six. On days one, three and five, the numbers of live, dead, and missing fish were scored for each replicate. Water temperature, pH, and dissolved oxygen (DO) were measured daily. Ammonia nitrogen (NH₃-N) was measured prior to each water renewal. At test termination, temperature, pH, DO, electric conductivity (EC), and ammonia were measured for each treatment. Overall, no significant deviations between measured water parameters among treatments or replicates were detected. The number of dead fish was counted at the end of the experiment (day 7) and surviving fish were sacrificed using an overdose of the anaesthetic MS-222 (Sigma, St Louis, MO, USA) in icewater to minimize degradation of RNA. Fork length (to nearest mm) and weight (to nearest 0.1 g) of each fish were recorded. No significant differences in length or weight were detected between individual treatment groups of the exposure experiments. Surviving individuals were sampled for subsequent analyses of sublethal biomarkers. Fifteen fish per treatment (three fish per replicate) were dissected immediately after individuals were sacrificed and measured. The entire gill apparatus, brain, liver, spleen, anterior kidney and two pieces of epaxial muscle from the left flank (< 30 mg) were removed, placed in sterile, RNase and DNase free 1.5 mL Eppendorf vials, and immediately snap-frozen in liquid nitrogen. Samples were stored at -80°C until RNA extraction and cDNA synthesis.

Quantitative real-time PCR: Frozen tissue samples (approximately 10 mg of liver, muscle and gill, brain, 9 mg total spleen and four mg total anterior kidney) were transferred to 1.5 mL collection tubes (RNeasy Mini Kit, Qiagen Inc., Valencia, CA), re-immersed in liquid nitrogen and ground to a fine powder with a sterile pestle. Subsequently, 350 µL of RNeasy lysis buffer (RLT, RNeasy Mini Kit, Qiagen Inc.) were added, and lysates were homogenized by pestle and by passing them through a pipette tip approximately 10 times. After incubation for three minutes at room temperature, the RNA was extracted according to the manufacturer's recommendations (RNeasy Mini Kit, Qiagen Inc.). Thereafter, 20 µL of each freshly extracted nucleic acid sample was digested with 10 U of RNase free DNase I (Roche, Mannheim, Germany) for 15 min at 37°C to remove genomic DNA. DNase digested RNA was quality controlled for absence of genomic DNA contamination. All samples had a minimal difference of 7 CT values between the cDNA and digested total RNA (tRNA), indicating that remaining gDNA contamination in the tRNA was 1% or less. Complementary DNA (cDNA) was synthesized using 100 units of SuperScript III (Invitrogen, Carlsbad, CA, USA), 600 ng random hexadeoxyribonucleotide (pd(N)6) primers (random hexamer primer), 10 U RNaseOut (RNase inhibitor), and 1 mM dNTPs (all Invitrogen, Carlsbad, CA, USA) in a final volume of 40 µL. The reverse transcription reaction proceeded for 50 min at 50°C. After addition of 60 µL of water, the reaction was terminated by heating for five min to 95°C and cooling on ice.

A suite of real-time TaqMan PCR systems for proteotoxicity (HSP70, HSP90), phase I detoxification mechanism (CYP1A1), metal-binding (metallothionein), endocrine disruption (vitellogenin), immune-system functioning and pathogen-defense (TGF-B, Mx-protein, nRAMP) were used based on Geist et al. (2007) for studying sublethal stress response at the transcriptome

level. L9 was quantified as internal reference. Real-time TaqMan PCR mixes contained 400 nM of each of two primers and 80 nM of the appropriate TaqMan probe. We used TaqMan Universal PCR Mastermix (Applied Biosystems, Foster City, CA, USA) containing 10 mM Tris-HCl (pH 8.3), 50 mM KCl, 5 mM MgCl₂, 2.5 mM deoxynucleotide triphosphates, 0.625 U AmpliTaq Gold DNA polymerase per reaction, 0.25 U AmpErase UNG per reaction and 5 µL of the diluted cDNA sample in a final volume of 12 µL. The samples were placed in 384 well plates and cDNA was amplified in an automated fluorometer (ABI PRISM 7900 Sequence Detection System, Applied Biosystems). Amplification conditions were two min at 50°C, 10 min at 95°C, 40 cycles of 15 s at 95°C and 60 s at 60°C. Fluorescence of samples was measured every 7 s and signals were considered positive if fluorescence intensity exceeded 10 times the standard deviation of the baseline fluorescence (threshold cycle, C_T). SDS 2.2.1 software (Applied Biosystems) was used to quantify transcription.

Relative quantification of stress response gene transcription: The comparative C_T method was applied to quantify gene transcription of investigated stress response genes (User Bulletin #2, Applied Biosystems). Values are reported as relative transcription or the n-fold difference relative to a calibrator cDNA (i.e. average target gene transcription of control fish). Three housekeeping genes (18S, L9, GAPDH) were tested and the one revealing smallest standard deviation and most stable transcription levels over all treatments (L9) was used to normalize the target gene signals (ΔC_T) for the differences in the amount of nucleic acid added to each reaction and the efficiency of the reverse transcriptase step. The ΔC_T for each experimental sample from the exposed fish was subtracted from the ΔC_T of the calibrator, the mean target gene signal of control fish. Finally, the linear amount of target molecules relative to the calibrator was calculated by $2^{-\Delta\Delta C_T}$. Therefore, all stress response gene transcriptions are expressed as an n-fold difference relative to the calibrator. For comparisons of basic linearized transcription values between tissues of all pooled control fish, muscle tissue revealed lowest transcription levels in all stress response genes and average transcription of each stress response gene in muscle was thus used as a calibrator for other tissues.

Statistical Analyses: Gene transcription data were first tested for normality and equality of variances. Since more than the randomly expected number of data sets was either not normally distributed or failed equality of variance tests, we generally used non-parametric methods for comparisons between treatments and tissues. Kruskal-Wallis One Way Analysis of Variance on Ranks (K-W ANOVA) was used to detect differences in linearized mean responses between treatments and tissues. In case of significance ($p < 0.05$), we tested for (i) differences in gene transcription between control and treatment groups and (ii) differences in gene transcription between the tissue with the weakest transcription level and other tissues by using non-parametric Mann-Whitney U-test. For comparisons between tissue types, Bonferroni corrections were applied to adjust p-values for multiple comparisons. We decided to use a conservative and non-parametric statistical approach throughout the dataset for simplicity and in order to reduce the number of false-positives. It should be noted, however, that the robustness of data interpretation is strengthened by the fact that these results were very similar to those obtained by using parametric tests (One Way Analysis of Variance, ANOVA and Dunn's or Tukey's Post-Hoc tests) with the limitation that some comparisons could not have been carried out under the requirements for equality of variances and normal distribution. Statistical analyses were carried out using the statistical programs Statistica 6.0 (StatSoft Inc., Tulsa, OK, USA), SPSS 7.0 and

SigmaStat 2.0 (SPSS, Inc., Chicago, Illinois).

7.2.2.2 Results

Results of quantitative PCR analysis for each tissue analyzed are shown in Table 7-6. While temperature (20°C), pH (7.17-7.88), dissolve oxygen concentration (7.9-9.2 mg/L) during the 7-d test varied little between treatments, but the EC/salinity showed a wide range of values across sampling sites. Thus, results obtained for site 340 with an EC of 16,070 µS/cm at 20°C need to be compared to the “high-EC control” (EC=18,800 µS/cm at 20°C). Sites 508 and 609 had ECs of 3007 and 4887 µS/cm at 20°C, respectively, and were thus compared to both control and high-EC control. Sites 711, 910 and 915 were compared to the control only.

Table 7-6: Changes in stress-response gene expressions in the liver, brain, kidney, gills, spleen and muscle of the striped bass (*Morone saxatilis*) exposed to Delta water samples collected on August 22/23, 2006. Results are presented as n-fold linear differences to ribosomal-L9 control gene expression with respective standard errors (S.E.). Boxed data containing the symbols *, ** and *** refer to significant differences to High EC (black) and Reference (blue) controls, at *p*-values of <0.05, <0.01 and <0.001 respectively; Kruskal-Wallis analysis of variance (GraphPadPrism 5.01). Outliers were removed using Grubb’s Test; extreme studentized deviate methods.

LIVER

		High EC	Control (Ref)	508	609	915	910	711	340
HSP 70	Mean	2.12	0.07	2.18	0.30	0.28	3.03	-1.19	3.06
	S.E.	0.76	0.63	1.02	0.16	0.65	1.07	0.47	1.25
HSP 90	Mean	1.27	-0.11	1.31	0.93	0.35	3.46	-0.90	3.60
	S.E.	0.67	0.71	0.89	0.59	0.65	1.36	0.53	1.23
CYP 1a	Mean	5.84	-0.04	2.69	2.22	1.51	6.88	0.15	9.31
	S.E.	2.12	0.69	1.53	0.88	0.83	2.07	0.50	2.58
TGF-B	Mean	-8.72	-0.74	-2.75	1.10	0.85	-5.68	-0.03	2.51
	S.E.	8.87	1.93	1.49	0.31	0.57	3.66	0.41	0.51
MT	Mean	3.27	0.12	2.39	0.90	3.47	4.36	1.85	2.70
	S.E.	1.15	0.94	2.12	0.25	0.75	1.47	0.53	1.43
MX	Mean	0.76	0.52	-0.36	1.22	6.88	14.10	0.76	24.74
	S.E.	3.60	1.55	1.34	0.32	3.91	6.04	1.89	8.85
nRAMP	Mean	-9.14	-0.26	0.86	3.29	4.41	-6.40	1.69	2.40
	S.E.	3.67	1.13	1.11	0.22	1.77	5.33	0.50	1.39
Vtg	Mean	-156.26	-66.82	-276.28	-326.20	-130.42	-76.39	-252.41	-169.14
	S.E.	47.22	206.53	107.04	6.88	37.24	19.50	35.81	45.68

BRAIN

		High EC	Control (Ref)	508	609	915	910	711	340
HSP 70	Mean	2.04	0.06	-0.87	6.30	-1.97	-4.95	-2.03	-2.63
	S.E.	1.05	0.58	1.10	5.78	1.38	3.08	0.78	1.27
HSP 90	Mean	0.35	-0.70	-4.93	0.27	-9.32	-20.14	-5.33	-4.98
	S.E.	0.69	1.27	2.29	0.94	3.68	** 6.40	3.15	2.95
CYP 1a	Mean	-0.13	-0.34	565.77	1.64	2.38	-3.40	-43.07	426.37
	S.E.	1.22	0.85	345.70	4.07	2.94	3.82	40.53	210.63
TGF-B	Mean	386.94	*** -0.28	55.44	** 240.07	76.64	1.40	* 44.37	43.71
	S.E.	155.39	0.47	* 14.49	** 238.53	* 32.48	0.50	20.68	9.19
MT	Mean	-487.34	*** -0.60	-43.84	** -80.34	-436.06	-21.99	* -513.27	-176.01
	S.E.	137.40	1.31	41.45	80.06	*** 229.45	15.84	352.22	37.66
MX	Mean	21.18	*** -0.42	58.01	* 14.58	23.27	-1.63	* 2.05	5.09
	S.E.	5.58	1.47	22.13	15.62	20.35	1.34	1.43	1.03
nRAMP	Mean	-2.29	0.30	7.73	-2.88	-4.37	-5.94	-5.25	-3.10
	S.E.	0.62	0.52	3.47	* 0.08	* 1.32	2.88	** 1.47	1.70

KIDNEY

		High EC	Control (Ref)	508	609	915	910	711	340
HSP 70	Mean	0.63	-0.31	-0.88	0.11	2.47	1.29	-0.62	1.42
	S.E.	0.55	0.39	0.34	0.74	1.05	0.36	0.48	0.87
HSP 90	Mean	0.76	-0.04	-0.60	-0.13	4.91	0.11	0.75	2.13
	S.E.	0.58	0.58	0.37	0.78	2.65	0.43	0.57	0.87
CYP 1a	Mean	3.18	0.03	-5.32	37.53	22.44	13.60	39.64	13.38
	S.E.	1.16	0.51	6.77	*** 18.68	9.49	6.55	*** 12.73	4.25
TGF-B	Mean	-0.63	0.35	-0.51	*** 6.94	5.67	2.06	5.66	0.57
	S.E.	0.35	0.39	0.47	* 2.17	1.79	1.67	1.10	0.51
MT	Mean	-0.31	-0.20	-0.46	-51.56	0.03	-3.44	-12.48	-4.35
	S.E.	0.87	1.12	0.90	51.19	3.01	0.93	** 3.02	0.83
MX	Mean	1.27	1.35	1.63	** 82.53	75.14	43.57	31.14	4.38
	S.E.	1.53	2.12	1.63	*** 46.24	36.53	21.99	*** 6.98	0.77
nRAMP	Mean	0.55	1.47	-0.17	3.97	3.54	1.85	4.26	-3.18
	S.E.	0.40	2.28	0.36	*** 2.11	0.93	0.95	*** 0.85	4.17

GILLS

		High EC	Control (Ref)	508	609	915	910	711	340
HSP 70	Mean	8.91	*** -0.03	5.16	20.50	3.17	8.01	35.24	5.27
	S.E.	2.24	0.43	3.28	*** 13.49	0.59	* 3.08	*** 17.95	0.93
HSP 90	Mean	10.21	*** -0.15	2.93	8.08	3.38	7.51	52.99	5.48
	S.E.	2.92	0.51	3.20	*** 0.77	0.56	2.92	*** 29.33	1.11
CYP 1a	Mean	10.34	0.04	35.59	53.04	9.35	25.49	109.79	19.35
	S.E.	3.01	0.54	*** 14.05	*** 26.23	2.83	* 11.91	*** 65.34	4.02
TGF-B	Mean	-29.51	0.03	-10.57	* -4.08	0.64	2.28	-3.07	-2.13
	S.E.	22.40	0.55	24.50	* 8.07	1.01	* 1.01	2.25	4.59
MT	Mean	13.43	*** 0.09	-171.98	12.30	3.78	18.62	134.49	5.44
	S.E.	4.13	0.42	* 168.27	* 7.07	0.82	*** 8.69	*** 76.08	1.28
MX	Mean	8.88	-0.10	25.42	6.25	11.91	38.10	6.11	9.76
	S.E.	2.65	0.49	13.66	0.74	5.23	*** 10.11	4.73	2.11
nRAMP	Mean	-13.86	-0.05	15.19	2.72	-1.81	1.01	-8.69	-3.73
	S.E.	14.06	0.45	7.50	1.55	3.18	1.11	4.66	1.25

SPLEEN

		High EC	Control (Ref)	508	609	915	910	711	340
HSP 70	Mean	0.72	0.50	-2.91	-1.02	-1.42	-1.32	-1.40	-0.85
	S.E.	0.92	1.33	0.93	0.44	0.21	0.19	0.21	0.41
HSP 90	Mean	4.56	* 0.23	*** -1.65	1.32	1.20	0.51	0.98	2.10
	S.E.	2.14	0.60	0.25	0.15	0.39	0.37	0.29	0.60
CYP 1a	Mean	-0.93	3.04	-7.16	1.64	0.19	-4.39	-16.64	11.31
	S.E.	2.63	4.22	4.16	4.44	2.24	5.08	4.77	7.13
TGF-B	Mean	1.22	0.20	0.46	1.08	1.72	-0.24	-0.46	0.90
	S.E.	1.13	0.67	0.42	0.63	0.20	0.38	0.36	0.47
MT	Mean	2.14	2.30	*** -5.68	** -3.09	-2.65	-1.27	-0.10	-0.74
	S.E.	0.76	3.25	0.78	2.30	0.99	0.83	0.62	0.61
MX	Mean	3.38	-0.21	-1.10	0.37	6.34	4.46	-1.01	5.56
	S.E.	2.21	0.81	0.42	0.57	3.13	1.22	0.78	0.89
nRAMP	Mean	-0.70	0.14	0.31	-0.22	0.70	2.04	0.52	0.25
	S.E.	0.96	0.65	0.43	0.15	0.34	0.20	0.45	0.44

MUSCLE

		High EC	Control (Ref)	508	609	915	910	711	340
HSP 70	Mean	-0.46	-0.13	-0.98	0.27	-0.33	-2.00	0.79	1.81
	S.E.	0.59	0.60	1.96	0.25	0.39	0.61	0.46	0.35
HSP 90	Mean	-1.10	-0.24	1.98	-0.82	-0.71	0.27	-1.22	0.81
	S.E.	1.33	1.00	7.01	0.52	0.54	0.77	0.44	0.41
CYP 1a	Mean	0.83	-0.02	2.49	1.86	-0.85	-0.79	1.41	5.81
	S.E.	1.07	1.30	0.26	2.38	0.59	0.65	1.22	1.59
TGF-B	Mean	0.13	0.10	0.31	-0.15	-0.01	-1.89	-0.82	1.77
	S.E.	0.58	0.73	7.94	1.19	0.60	0.43	0.41	0.82
MT	Mean	0.20	-0.30	4.96	-1.67	-1.86	2.23	-0.79	-1.43
	S.E.	1.60	1.03	3.23	0.80	0.89	3.04	1.58	1.11
MX	Mean	-0.66	0.04	4.25	-0.01	4.94	7.47	6.18	1.62
	S.E.	1.09	0.81	1.93	0.31	2.91	5.94	5.27	2.52
nRAMP	Mean	0.12	-0.02	-0.78	1.16	-0.40	-1.16	0.39	-8.16
	S.E.	0.56	0.63	0.77	0.97	0.46	0.45	0.58	3.70

Results of quantitative PCR analysis are summarized in Table 7-7. Brain and gill tissues showed the strongest differences in response to EC changes, and results for these tissues from fish exposed to water from sites 508 and 609, have to be treated with caution, since no direct comparison to low EC or high EC controls is possible. Cyp1A1 mRNA was significantly elevated in gill tissue at sites 508, 609, 910 and 711, and in kidney at sites 609 and 711. Stress proteins HSP70 and HSP90 were induced in gills at sites 609 and 711. The cytokines nRAMP and Mx-protein were upregulated in kidney at sites 609 and 711. Metallothionein (Mt) was upregulated in gills at sites 910 and 711. Further data analysis is ongoing.

Table 7-7. Summary of results of quantitative PCR analysis on striped bass tissues exposed to Delta water samples.

Tissue	Site 508	Site 609	Site 915	Site 910	Site 711	Site 340
Liver	-	nRAMP↑	-	Vg↓	-	Mx↑
Brain	-	-	TGF-b↑ Mt↓ nRAMP↓	Hsp90↓	nRAMP↓	-
Kidney	-	Cyp1A↑ TGF-b↑ Mx↑ nRAMP↑	-	-	Cyp1a↑ Mt↓ Mx↑ nRAMP↑	-
Gills	Cyp1A↑ Mt↓	Hsp70↑ Cyp1A↑	-	Hsp70↑ Cyp1A↑ TGF-b↑ Mt↑ Mx↑	Hsp70↑ Hsp90↑ Cyp1a↑ Mt↑	-
Spleen	Hsp90↓ Mt↓	Mt↓	-	-	-	-
Muscle	-	-	-	-	-	-

7.2.3 Tissue-Specific Expression of Stress Response Genes in Striped Bass Exposed to Extracts of Semi-Permeable Membrane Devices (SPMD) Deployed in the Sacramento-San Joaquin Delta

7.2.3.1 Methods

To assess the presence and effects of bioavailable lipophilic contaminants in the estuary Semi-Permeable Membrane Devices (SPMDs) were deployed in three locations in the Delta on August 16, 2005, and retrieved on September 13, 2005, by D. Ostrach, UC Davis. SPMDs bind nonionic organic compounds with K_{ow} 's >1 (in practice, a chemical's K_{ow} should be greater than 200) and some neutral organo-metal complexes (Table 7-8). One SPMD was deployed in the Napa River (Napa) just below the new bridge, a second device was placed off Collinsville attached to the Bureau of Reclamation pier (Collinsville) and the third device deployed in Sand Mound Slough where high concentrations of *Microcystis aeruginosa* have been recorded (Sand Mound).

Upon retrieval, the devices were frozen and sent to Environmental Sampling Technologies (EST) Labs Inc. (<http://www.est-lab.com>) for processing within 24 hours. Extracts were then submitted to the California Department of Fish and Games Wildlife Water Pollution Control Laboratory, Rancho Cordova, CA, for chemical analyses and to process SPMD extracts for use in fish injection experiments. Results from the chemical analysis of the SPMDs indicated the presence of elevated levels of polycyclic aromatic hydrocarbons (PAHs) at all three Delta sites (see POD report to IEP by D. Ostrach, UC Davis) during the deployment period (mid-August to mid-September 2005).

Table 7-8. Common contaminates bound by SPMDs.

Acronym	Name	Possible Source
PAH	Polycyclic Aromatic Hydrocarbons	Combustion by-product
OC	OrganoChlorides	Pesticide
PCB	Polychlorinated Biphenyls	Industrial and electrical
	Pyrethroids	Insecticide
	Dioxins	Combustion, industrial
	Furans	Industrial by-product
	Nonyl Phenols	Industrial
	Alkylated Selenide	Fossil fuels

On March 6, 2006 the SPMD extracts (100 µL/fish) were injected into juvenile striped bass (10 fish/treatment) in an attempt to determine potential effects of the bioavailable, lipophilic contaminants on juvenile striped bass (see POD Report to IEP, D. Ostrach). In addition to injecting SPMD extracts several negative and positive control treatments were run concurrently using the same injection volume of 100 µL/fish: an unhandled control, peanut oil-only injection (carrier control), a dialysis blank (method blank), field blank (SPMD device opened at the field site during deployment then extracted to control for atmospheric contamination), beta-naphthoflavone at a dose of 2.5 mg/kg (positive control for P4501A1 induction), estradiol at a dose of 3.0 mg per kg (positive control for exposure to estrogenic compounds) and chlorpyrifos at a dose of 0.5 mg per kilogram (positive control for AChE inhibition). The experiment was terminated on March 9, 2006. Only 1 of 100 fish died during the experiment. Upon termination of the experiment fish were euthanized with MS-222, dissected and organs snap-frozen in liquid nitrogen and stored at -80°C for biochemical and molecular assays. Spleen and liver samples were analyzed for molecular biomarkers following methods described in Chapter 7.2.2.

7.2.3.2 Results

Changes in the expression of stress-response genes in the liver and spleen of juvenile striped bass (*M. saxatilis*) are presented in Table 7-9 below. Control treatments (oil only, non-handled, field blank) did not induce any of the stress response genes quantified here. The positive control for estrogen-inducing chemicals (estradiol) significantly increased vitellogenin transcription in the liver. The “dialysis blank” did increase transcription of Mt in the liver and Cyp1a in the spleen. SPMD extracts from all three field sites produced gene responses in the liver, but not the spleen, of exposed fish. Extract from the Collinsville site down-regulated transcription of Cyp1a and Mt, while extracts from Sand Mound and Napa down-regulated transcription of Mt only. Vitellogenin was slightly increased in fish exposed to SPMD extracts from Collinsville. Further analysis of the data is ongoing.

Table 7-9. Changes in gene expression of stress response genes as n-fold linear differences to ribosomal-L9 control gene expression with respective standard errors (S.E.). ** and *** refer to significant differences to oil injected controls, at *p*-values of <0.01 and <0.001 respectively; Kruskal-Wallis analysis of variance (GraphPadPrism 5.01).

		Oil only	Non-handled	BNF	Chlorpyrifos	Estradiol	Field Blank	Dialysis Blank	Collinsville	Sand Mound	Napa
		Controls		SPMD/Chemical Controls					Field Samples		
LIVER											
HSP 70	Mean	-4.0	0.1	-8.4	-4.8	-10.4	-9.8	-16.8	-2.0	5.7	-23.0
	S.E	2.8	0.6	4.5	2.4	4.2	3.4	5.2	1.3	1.7	10.7
HSP 90	Mean	-1.4	0.1	-4.8	-1.7	-7.9	-4.0	-11.0	-0.8	7.1	-7.5
	S.E	1.5	0.6	1.5	1.8	3.7	1.3	3.9	1.4	2.9	4.0
CYP 1a	Mean	0.8	0.0	1.6	-0.8	-2.9	-1.1	-3.4	-1719.2***	-241.6	-4.6
	S.E	2.7	0.7	0.8	0.9	0.6	0.6	0.6	569.1	158.5	1.3
TGF-B	Mean	-1.1	0.3	-1.2	-0.7	-2.2	-2.9	-7.4	45.2	1159.7	204.4
	S.E	1.5	0.5	1.6	1.1	1.8	1.6	3.8	28.3	451.4	212.0
MT	Mean	2.6	-0.1	-7.1	-3.7	-2.3	-2.8	-7.7**	-129.0***	-163.3***	-17.1**
	S.E	0.9	0.7	4.4	2.2	1.3	1.3	1.7	51.9	97.6	7.4
MX	Mean	1.7	-0.1	-3.6	-2.7	-0.5	-2.5	-1.7	29.0	69.5	3.6
	S.E	0.9	0.8	3.3	1.7	1.8	1.2	1.5	10.6	14.3	12.2
nRAMP	Mean	2.8	0.0	-0.4	0.2	7.6	1.4	-6.9	6.6	71.7	-3.1
	S.E	0.7	0.6	1.5	1.0	2.0	2.0	3.7	1.9	24.7	14.8
Vtg	Mean	5.0	-0.5	-	-	229392.7***	10.9	3.0	43.6	2.1	2.6
	S.E	3.6	1.5	-	-	73611.1	7.3	1.4	20.8	0.9	1.5
SPLEEN											
HSP 70	Mean	0.6	0.0	0.5	1.1	2.4	2.4	1.0	0.4	-0.9	-0.9
	S.E	0.6	0.7	0.4	0.5	0.3	0.4	0.7	0.6	0.5	0.8
HSP 90	Mean	0.1	-0.1	0.1	-0.5	4.0	3.1	-1.3	0.7	-1.0	-1.5
	S.E	0.7	1.0	0.4	2.0	3.2	0.7	0.6	0.8	0.6	1.1
CYP 1a	Mean	7.3	1.1	6.3	1.1	0.3	3.0	2197.5	6.1	5.7	0.3
	S.E	4.7	2.7	1.3	0.8	0.8	2.0	959.0	2.2	8.0	1.4
TGF-B	Mean	-0.4	-0.1	-0.6	0.1	3.8	2.9	3.0	0.6	-1.5	-0.8
	S.E	0.7	0.5	0.4	0.6	2.0	0.9	1.0	0.7	0.5	0.7
MT	Mean	3.2	-0.1	2.6	3.5	8.8	11.9	-7.6	10.0	2.6	-1.9
	S.E	1.0	1.0	0.6	0.8	5.9	4.2	12.0	6.5	0.7	4.0
MX	Mean	1.1	-0.1	1.7	0.4	0.9	2.9	1.9	0.2	1.6	0.4
	S.E	1.2	0.7	0.4	0.6	4.3	1.1	2.1	0.7	1.5	1.9
nRAMP	Mean	-0.3	0.0	-2.2	-0.9	2.7	2.1	0.2	0.4	-1.6	-3.0
	S.E	0.6	0.5	0.4	0.4	1.4	0.7	0.9	0.5	0.6	1.8

7.3 Identification of Molecular Biomarkers in the Delta Smelt (*Hypomesus transpacificus*) Using Microarray Technology.

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In order to understand the effects of contaminants upon *Hypomesus transpacificus* we have constructed a microarray with over 8,000 Expressed Sequence Tags (ESTs). We applied this tool to measure gene responses on 60-day old juveniles exposed to 50 µg/L copper for 7 days. The sublethal effects of copper exposure in the delta smelt appear to be on neuro-muscular activity, respiration and metabolism, and we have identified a number of affected genes involved in cardio-muscular contraction, neuro-transmission, oxidative stress, metal ion binding, immunity and systemic inflammation, and digestion. Amongst the responding genes there was a significant up-regulation of osteonectin, a source of copper-binding peptides, which may be indicative of tissue damage caused by excess copper. Future work will include further microarray analyses of delta smelt exposed to different toxicants, and investigation of a selected suite of genes from these microarray assessments, using real-time quantitative PCR to develop informative molecular biomarkers of stress and exposure in the delta smelt.

7.3.1 Introduction

The Delta smelt (*H. transpacificus*) is a pelagic fish species endemic to the Northern Sacramento-San Joaquin Estuary, California, and considered an “indicator species” for ecosystem health in this system. Abundance has dramatically declined since the 1980s and it was listed as threatened in 1993, under both the Federal Endangered Species Act (ESA) and California Endangered Species Act (CESA). Delta smelt have been reared since 1992 at the Fish Conservation and Culture Laboratory (FCCL), UC Davis, providing a refuge population as well as a supply for research. A more recent step decline of the delta smelt population (Sommer et al.

2007) has prompted considerable efforts to understand the causative factors of this decline. A number of complex factors, known and unknown have potentially been affecting populations of delta smelt in its native habitat. Pollution, in the form of agricultural, pharmaceutical and industrial chemicals, along with the effects of water exports for agricultural irrigation and urban uses, toxic algal blooms and habitat destruction, are among the potential causes for the decline in pelagic organisms.

Identifying the impacts of such stressors and their mechanistic effects on individuals and populations is a main challenge in ecotoxicology. Stress responses to toxic chemicals are often preceded by alterations in gene expression, thus gene expression studies offer insights into the overall health of an organism. Microarray gene profiling is a powerful tool for defining genome-wide effects of environmental change on biological function. This technology is being applied successfully to the field of ecotoxicology in a number of other species and links are being forged between what is measured at the gene expression level and life history parameters, such as metabolism, growth and reproduction (Connon et al, 2008, Heckmann et al, 2008). The predictive value of microarrays as screening tools is becoming more powerful as our understanding of these responses grows. Gene expression studies carried out over short-term exposures allow for the prediction of chronic effects that stressors may have on the health of the individual, their survival capacity, fecundity and somatic growth. Specific gene responses in individual delta smelt, indicative of their health status, could highlight potential causes for the population decline.

Our aims are to determine specific and general responses to a suite of stressors and develop molecular biomarkers applicable in the delta smelt and relevant to the varying contaminants found in the Californian watersheds. In order to understand the effects of contaminants upon *H. transpacificus* we have constructed a microarray with over 8,000 Expressed Sequence Tags (ESTs). No sequence information was available on any database at the time this project was started. We describe here, the construction and first application of this tool to measure gene responses to copper in juvenile delta smelt. We used copper to generate stress because biochemical responses to this metal, and adverse effects on the whole organism are relatively well understood and therefore would aid interpretation of results in this “proof of principle” test. Furthermore, copper is a contaminant of concern in Californian waterways, it is a common contaminant in urban storm-water runoff, is present from mining activities and is regularly used as a pesticide in agricultural areas. We expect neurological responses, respiration, growth and metabolism to be affected by exposure to this neurotoxin. We investigate relatively high levels of copper ($50\mu\text{gCu}^+\text{L}^{-1}$) in order to establish confidence in significant responses. Reported concentrations of copper in the Sacramento River are above $6\mu\text{g Cu}^+\text{L}^{-1}$ (USGS, 1998) though there are seasonal fluctuations due to its application as a pesticide, where concentrations have been reported to exceed $500\mu\text{g Cu}^+\text{L}^{-1}$ in rice field effluents, following copper application (Department of Fish and Game, California, 1998).

7.3.2 Methods

Microarray construction and hybridization. We constructed a delta smelt microarray using 8448 PCR amplified fragments from a normalized cDNA library. To ensure presence of

potential genes of interest, in the construction of this tool, we used organisms exposed to a range of conditions/stressors, listed in Table 7-9. Total RNA was extracted from treated fish and specific organs using a Qiagen RNeasy kit according to manufacturer's protocols and pooled into a single sample that was used to construct a cDNA library for expressed sequence tags (ESTs) ligated to *p-bluescript* plasmid vectors and cloned into chemically competent *Escherichia coli* cells (BioS&T Inc, Montreal, Quebec, Canada). Aliquots from the cDNA library were cultured overnight at 37°C, on nutrient agar plates containing 100 µg X-gal/L and 100mM isopropyl β-galactosidase (IPTG) for blue-white screening. White colonies were picked using sterilized toothpicks and individually cultured in 100µl Luria Bertani (LB) media for 4 hours at 37°C, in flat-base 96-well plates and stored in 15% glycerol at -80°C for subsequent amplification.

Table 7-9. List of stressors and treatments delta smelt were exposed to, from which RNA was extracted for the construction of a cDNA library.

Water Sample or Stressor	Tissue	Age
Groundwater	Whole fish	10-day old
SWAMP	Whole fish	10-day old
CDM	Whole fish	10-day old
Low salinity (159 µS.cm ⁻¹)	Whole fish	10-day old
High salinity (3630 µS.cm ⁻¹)	Whole fish	10-day old
Temperature 20°C. Hatchery water	Whole fish	10-day old
Esfenvalerate (0.125 µg/L)	Whole fish	10-day old
Copper (25 µg/L)	Spleen	60-day old
Copper (25 µg/L)	Brain	60-day old
Copper (25 µg/L)	Muscle	60-day old
Copper (25 µg/L)	Gonad	60-day old
Copper (25 µg/L)	Liver	60-day old
Copper (25 µg/L)	Whole fish	60-day old
Site 915	Whole fish	90-day old
Site 711	Whole fish	90-day old

A total of 8,448 ESTs (88 x 96-well plates) were PCR amplified directly from the bacterial colonies, using 1 μ l bacterial suspension with M13 long primers (MWG Biotech):

- M13 rev (-49) 5'-GAGCGGATAACAATTTTCACACAGG-3'
- M13 uni (-43 5'-AGGGTTTTCCAGTCACGACGTT-3'

Following a cycling program with an initial denaturation of 95°C for 15 min, 35 cycles of denaturation at 95°C for 45 sec, primer annealing at 53°C for 30 sec and elongation at 72°C for 3 min, followed by a final extension at 72°C for 10 min. Amplified PCR products were visualized on agarose gels and ranged in size from 1- 4kb. Products were vacuum purified using Minelute 96 UF PCR Purification System (Qiagen) as per manufacturers' protocol and transferred to 22 x 384-well plates. Plates were desiccated using a vacuum centrifuge and products resuspended at concentrations between 0.1-0.5 μ M required for printing, in a 1x phosphate buffer solution (Nexterion).

PCR fragments and controls were pin-printed on glass slides in a 20 x 19 block format, with 48 blocks per microarray [Grid = 18,240 spots (8448 clones in duplicate = 16,895, plus 576 control spots (1,152 control) and 96 (192 blank) blank spots, also printed in duplicate and repeated throughout the array in each block)]. Microarrays were printed at the Array Core facility at Robbins Hall, UC Davis (<http://array.ucdavis.edu/home/>). Microarray control spots included a number of hybridization tags comprised of a pooled PCR product from all spots on the array, *H. transpacificus* DNA, and four Spot Report System of alien PCR products from *Arabidopsis thaliana*; CAB, RCA, RBCL and LPT4 (Stratagene, USA). Blank control spots consisting of 1x Nexterion buffer solution were printed interspaced with the above controls and as the last 12 spots in each block, and used to assess printing quality and potential cross contamination resulting from printing.

Fish Exposures. Procedures and methods for the copper exposure are described in Chapter 6.1.3.2. Briefly, fish obtained from the Fish Conservation and Culture Laboratory, UC Davis, were exposed to a control and four concentrations of copper chloride (CuCl₂); equivalent to nominal concentrations of 5, 10, 25 and 50 μ g Cu⁺ L⁻¹ for 7 days. Replicate experimental treatment (n=4) were initiated with 10, 60-day old juveniles in 7L of water at 20°C. Fish were fed twice daily with artemia (<48 h old). The light:dark cycle was 16h:8h. Approximately 80 percent of the water in each replicate was renewed on days 2, 4, and 6. On days 1, 3, and 5, water was not renewed, but the numbers of live, dead, and missing fish were scored for each replicate. Water temperature, pH, and DO were measured daily. Ammonia nitrogen (NH₃-N) was measured prior to each water renewal. On Day 7, 4 fish from each replicate were measured for mass and fork length recorded for surviving fish prior to snap-freezing and storage at -80°C for subsequent analyses. Only controls and the highest exposure concentration (50 μ g Cu⁺ L⁻¹) were assessed with the microarray.

RNA isolation, cDNA synthesis and fluorescence labeling. RNA was extracted using a standard phenol:chloroform protocol with Trizol Reagent (Invitrogen). Fifteen micrograms of total RNA was used cDNA synthesis, spiked with control RNA (CAB, RCA, rbcl and LTP4 (SpotReport, Stratagene) and labeled with Alexa fluor dyes, using SuperScript[™] Plus Indirect cDNA labeling System (Invitrogen). The fluorescently labeled probes were purified using QIAGEN PCR "Qiaquick" columns according to the manufacturer's instructions, and were

quantified spectrophotometrically (NanoDrop) to determine labeled cDNA concentration and dye incorporation. RNA from stock unexposed fish samples were similarly extracted and pooled to create a reference pool against which all samples would be hybridized. Experimental samples and control cDNA were labeled with Alexa fluor 647, and reference pool cDNA with Alexa fluor 555. No dye swaps were carried out as labeling was consistent throughout the reference design. Each experimental sample and control was combined with a reference pool cDNA prior to hybridization using an automated Tecan HS4800 hybridization station at 45°C. Slides were scanned using a GenePix 4000A scanner (Axon Instruments).

Microarray Analysis. Normalization and analytical methods are described in Loguinov *et al.* (2004). Print tip normalization was carried out within slides and sequential single slide data analysis was carried out as an alternative to between-slide normalization. An outlier-generating model was used to identify differentially expressed genes.

7.3.3 Results and Discussion

Water quality. Water chemistry remained stable throughout the 7-day exposure except for ammonia at the highest concentrations. However this was attributed to high mortality, and therefore lower number of fish remaining in these samples (Table 7-10).

Table 7-10. Summary of water chemistry measurements taken on termination of the delta smelt Cu⁺ reference toxicant test.

Treatment	Lab Temp (°C)	Lab pH	Lab EC (µmhos/cm)	Lab DO (mg/L)	Ammonia (mg/L)
Lab. Control (Dilute Well Water)	21	8.4	431	8.8	0.28
5 ppb Cu ⁺	21	8.49	456	8.7	0.24
10 ppb Cu ⁺	21	8.48	461	9	0.23
25 ppb Cu ⁺	21	8.46	455	8.8	0.37
50 ppb Cu ⁺	21	8.39	457	8.9	0.14

Toxicity test. The calculated EC_{50-96h} was 33.5µg.Cu⁺.L⁻¹ and EC_{50-7day} was 24.7µg.Cu⁺.L⁻¹ (Table 7-11 and Figure 7-1). The LC_{50s} of juvenile delta smelt for copper are far below the 96-h LC₅₀ value reported by the California Department of Fish and Game of 1.4 mg/L for larval delta smelt (Werner *et al.* 2005). Our experimental results and other available data indicate that delta smelt is one of the most sensitive fish species to copper. No significant differences were observed in length and weight after the 7-d exposure, though slight weight increase was observed at the higher concentrations attributed to fewer surviving organisms resulting in a relative increase of food and space compared to controls.

Microarray responses. Differentially expressed genes resulting from copper exposure are presented in Table 7-12 and categorized in Figure 7-2. Responses include involvement in cardiac muscular contraction, activity and neurological responses involved in calcium and

phosphate signaling. Digestion was also affected by copper, not only in the production of enzymes involved in food digestion but also specific to chitin (invertebrate) breakdown.

Table 7-11. Summary of 7-day delta smelt Cu⁺ reference toxicant test conducted using dilute well water spiked with copper chloride. Highlighted areas indicate a significant reduction in survival (p<0.05).

Treatment	Survival (%) ²		Length (cm) ²		Weight (g) ²	
	x	se	x	se	x	se
Laboratory Control (Dilute Well Water)	100.0	0.0	3.42	0.04	0.24	0.00
5 ppb Cu ⁺	93	6.7	3.53	0.05	0.24	0.01
10 ppb Cu ⁺	95	2.9	3.49	0.04	0.23	0.00
25 ppb Cu ⁺	40	4.1	3.57	0.11	0.26	0.02
50 ppb Cu ⁺	23	4.7	3.52	0.08	0.26	0.02

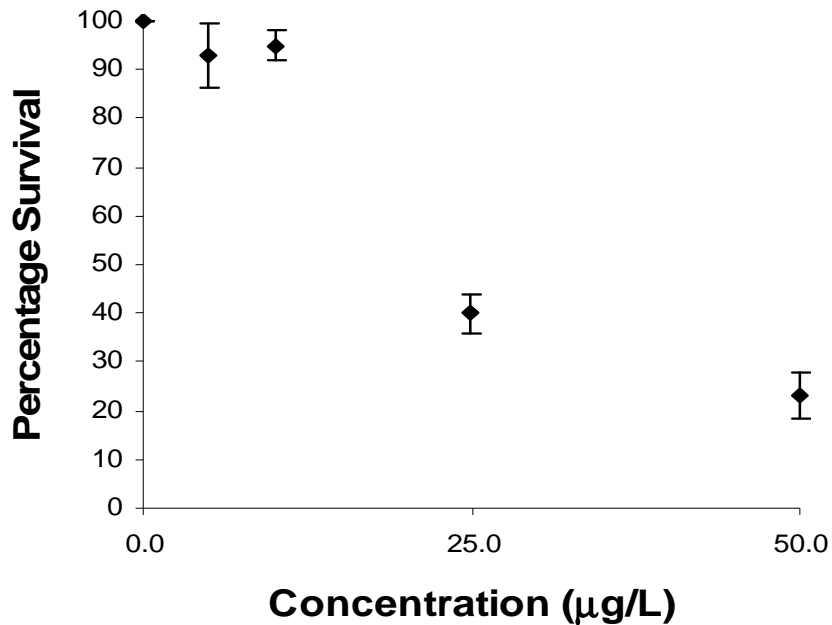


Figure 7-1. Delta smelt copper toxicity test. Percentage survival following 7-day exposure.

Table 7-12. Annotation and respective list of genes significantly responding to 7-day copper exposure (50 µg/L).

Small Clone ID	Response	Fold	Gene most similar to	Species Match	Accession No	E-Value	Score	GO	Gene Ontology Description
DS1961_39_B_06	Up	2.36	1-acylglycerol-3-phosphate O-acyltransferase 3	Danio rerio	NP_958920	4.00E-68	261	GO:0003841	1-acylglycerol-3-phosphate O-acyltransferase activity
DS1961_77_C_03	Up	4.88	actin, alpha 2, skeletal muscle	Pagrus major	BAF80060	1.00E-94	384	GO:0003774	motor activity
DS1961_03_E_12	Up	3.75	actin, alpha 2, smooth muscle, aorta	Danio rerio	AAH75896	e-107	391	GO:0003774	motor activity
DS1961_19_B_07	Up	2.92	actin, alpha, cardiac muscle 1 like	Danio rerio	NP_001001409	e-141	503	GO:0003774	motor activity
DS1961_34_A_06	Up	3.11	actin, alpha, cardiac muscle 1 like	Danio rerio	NP_001001409	e-148	529	GO:0003774	motor activity
DS1961_29_C_10	Up	3.28	actin, alpha, cardiac muscle 1 like	Danio rerio	NP_001001409	e-145	519	GO:0003774	motor activity
DS1961_12_G_03	Up	4.58	actin, alpha, cardiac muscle 1 like	Danio rerio	NP_001001409	e-125	449	GO:0003774	motor activity
DS1961_21_H_11	Up	6.10	actin, alpha, cardiac muscle 1 like	Danio rerio	NP_001001409	e-127	458	GO:0003774	motor activity
DS1961_28_C_06	Up	2.51	actin, beta	Acanthopagrus schlegelii	AAH84610	e-122	441	GO:0000586	cytoskeleton
DS1961_05_A_08	Up	1.83	acyl-CoA synthetase long-chain family member 5	Tetraodon nigroviridis	CAG06490	e-102	375	GO:0004467	long chain fatty acyl-CoA synthetase
DS1961_47_C_04	Up	3.47	aldolase A, fructose-bisphosphate	Danio rerio	NP_910358	e-124	447	GO:0006096	glycolysis
DS1961_63_E_08	Up	1.86	alpha tubulin, (protein LOC573122)	Danio rerio	NP_001008506	e-120	434	GO:0007018	microtubule-based movement
DS1961_05_B_06	Up	3.06	amylase-3 protein	Pseudopleuronectes americanus	AAE65927	e-144	513	GO:0004556	alpha-amylase activity
DS1961_70_B_05	Up	3.36	amylase-3 protein	Tetraodon nigroviridis	CAC87127	3.00E-54	213	GO:0004556	alpha-amylase activity
DS1961_01_C_12	Up	1.80	apolipoprotein	Tetraodon nigroviridis	CAG03661	1.00E-38	78	GO:0003001	cholesterol transport
DS1961_80_B_11	Up	2.28	apolipoprotein A-I	Danio rerio	NP_571203	1.00E-81	306	GO:0003344	cholesterol efflux
DS1961_74_G_02	Up	3.34	apolipoprotein A-I	Danio rerio	NP_571203	1.00E-81	306	GO:0003344	cholesterol efflux
DS1961_72_D_09	Up	7.47	apolipoprotein A-I	Danio rerio	NP_571203	6.00E-73	277	GO:0003344	cholesterol efflux
DS1961_11_H_09	Up	3.59	apolipoprotein A-I precursor (Apo-AI-1)	Oncorhynchus mykiss	057823	8.00E-76	286	GO:0003344	cholesterol efflux
DS1961_88_D_02	Up	4.81	apolipoprotein A-I precursor	Oncorhynchus mykiss	057524	4.00E-71	271	GO:0003344	cholesterol efflux
DS1961_02_A_11	Up	2.72	apolipoprotein A-IV	Danio rerio	AAH93239	1.00E-73	279	GO:0006869	lipid transport
DS1961_41_F_02	Up	2.23	apolipoprotein B	Salmo salar	CAA57449	3.00E-24	115	GO:0003001	cholesterol transport
DS1961_62_B_05	Up	2.02	apolipoprotein CII	Oncorhynchus mykiss	AAE11410	1.00E-19	100	GO:0006869	lipid transport
DS1961_65_E_08	Up	2.17	apolipoprotein CII	Oncorhynchus mykiss	AAE11410	3.00E-19	99	GO:0006869	lipid transport
DS1961_45_G_03	Up	4.16	apolipoprotein E2	Danio rerio	NP_571173	2.00E-38	162	GO:0003344	cholesterol efflux
DS1961_79_B_01	Up	4.42	arachidonate 12-lipoxygenase	Danio rerio	NP_958912	4.00E-23	112	GO:0004052	arachidonate 12-lipoxygenase activity
DS1961_77_B_08	Up	1.56	arachidonate 12-lipoxygenase	Danio rerio	NP_958912	5.00E-33	151	GO:0004052	arachidonate 12-lipoxygenase activity
DS1961_06_F_12	Up	3.07	atactin like metallo-protease	Oryzias latipes	NP_001080207	2.00E+03	311	GO:0005033	atactin activity
DS1961_17_B_07	Up	4.34	atactin like metallo-protease	Oryzias latipes	NP_001080207	7.00E-50	199	GO:0000853	atactin activity
DS1961_07_A_09	Up	2.17	atp-like protein	Acanthopagrus schlegelii	AAH94966	3.00E-38	162	GO:0004817	phosphate transport
DS1961_71_F_02	Up	3.27	calpain 1 protein	Danio rerio	AAH91999	2.00E-68	262	GO:0005509	calcium ion binding
DS1961_37_C_06	Up	1.82	carboxypeptidase H	Paralichthys olivaceus	AAO27852	1.00E-82	309	GO:0004183	carboxypeptidase H activity
DS1961_27_A_09	Up	2.91	cardiac muscle ATP synthase, alpha 1,	Danio rerio	NP_001070823	7.00E-62	240	GO:0001562	ATPase activity
DS1961_05_C_08	Up	5.76	cell division cycle 14 homolog A	Danio rerio	CAF09233	3.00E-19	99	GO:0004725	protein tyrosine phosphatase activity
DS1961_30_G_11	Up	2.34	chitin binding Peritrophin-A domain	Danio rerio	AAH45331	4.00E-69	244	GO:0016490	structural constituent of peritrophic membrane
DS1961_31_B_04	Up	3.75	chitinase	Oncorhynchus mykiss	CAB93487	9.00E-68	260	GO:0004867	chitinase activity
DS1961_69_C_08	Up	4.25	chitinase	Paralichthys olivaceus	BAB10559	e-127	458	GO:0004867	chitinase activity
DS1961_71_C_06	Up	3.93	chymotrypsinogen 2-like protein	Sparus aurata	AAE45254	1.00E-20	101	GO:0004263	chymotrypsin activity
DS1961_67_A_02	Up	5.20	chymotrypsin-lipotropin A precursor	Oncorhynchus mykiss	0Q4617	7.00E-63	244	GO:0005179	hormone activity
DS1961_11_H_03	Up	1.86	DAZAP2-like protein (deleted in azoospermia-associated)	Takifugu rubripes	NP_001072102	5.00E-59	230	GO:0030154	cell differentiation
DS1961_68_G_10	Up	5.08	elastase 2-like protein	Sparus aurata	AAE45251	2.00E-89	332	GO:0006508	proteolysis
DS1961_74_C_03	Up	3.73	F-type lectin	Morone saxatilis	ABE29997	1.00E-46	188	GO:0016467	proton-translocating F-type ATPase
DS1961_08_H_11	Up	2.93	gamma-synuclein	Takifugu rubripes	NP_001029019	2.00E-41	172	GO:0030424	axon
DS1961_71_D_02	Up	3.24	glutamate dehydrogenase 1	Danio rerio	NP_958939	e-107	392	GO:0004382	glutamate dehydrogenase activity
DS1961_70_G_02	Up	1.88	guanine nucleotide binding protein (G protein), beta 1	Danio rerio	NP_997774	e-117	424	GO:0000392	GTPase activity
DS1961_14_D_09	Up	3.76	hect domain and RLD 4 (similar to...)	Danio rerio	ZF_005685	7.00E-26	266	GO:0005312	ubiquitin cycle
DS1961_76_F_08	Up	3.02	hemopexin	Danio rerio	NP_001104617	1.00E-59	233	GO:0046872	metal ion binding
DS1961_37_H_01	Up	4.41	hemopexin	Danio rerio	NP_001104617	7.00E-94	347	GO:0046872	metal ion binding
DS1961_22_C_07	Up	3.62	histone methyltransferase Hmyb1b	Danio rerio	ABC54714	e-108	394	GO:0030239	myofibril assembly
DS1961_60_D_07	Up	2.82	histone fatty acid binding protein	Danio rerio	AAE09225	3.00E-56	221	GO:0000269	lipid binding
DS1961_33_F_03	Up	2.76	L-arginine:glycine amidinotransferase	Danio rerio	NP_958825	5.00E-83	310	GO:0016740	transferase activity
DS1961_36_H_12	Up	2.78	L-arginine:glycine amidinotransferase	Danio rerio	NP_958825	8.00E-91	316	GO:0016740	transferase activity
DS1961_04_E_11	Up	4.66	L-arginine:glycine amidinotransferase	Danio rerio	NP_958825	8.00E-92	340	GO:0016740	transferase activity
DS1961_83_E_10	Up	6.12	L-arginine:glycine amidinotransferase	Danio rerio	NP_958825	2.00E-08	328	GO:0016740	transferase activity
DS1961_65_G_04	Up	6.60	L-arginine:glycine amidinotransferase	Danio rerio	NP_958825	4.00E-95	340	GO:0016740	transferase activity
DS1961_25_A_10	Up	4.17	ligoxygase 12P (PF02182): similar to...	Oncorhynchus anatinus	ZF_00151817	8.00E-06	55	GO:0016165	lipoygase activity
DS1961_49_C_08	Up	2.21	muscle creatine kinase	Danio rerio	CAB16434	e-112	406	GO:0004111	creatine kinase activity
DS1961_77_G_08	Up	3.91	myosin II	NP_991241	2.00E-25	119	GO:0030346	protein phosphatase 2B binding	
DS1961_37_C_09	Up	3.47	NADH dehydrogenase subunit 5	Oreochromis mossambicus	AD135911	5.00E-94	300	GO:0000137	NADH dehydrogenase (ubiquinone) activity
DS1961_57_D_04	Up	3.88	NADH dehydrogenase subunit 5	Oreochromis mossambicus	AD135911	e-107	390	GO:0000137	NADH dehydrogenase (ubiquinone) activity
DS1961_44_B_02	Up	3.03	NADH dehydrogenase subunit 6	Selangichthys microdon	NP_798483	e-107	392	GO:0000137	NADH dehydrogenase (ubiquinone) activity
DS1961_03_E_05	Up	7.54	pancreatic protein with two somatomedin B domains	Paralichthys olivaceus	BAA80146	2.00E-95	352	GO:0005179	hormone activity
DS1961_12_F_09	Up	4.05	Pepsin A2	Tramotodus bernacchii	CAB06096	2.00E-08	253	GO:0004194	pepsin A activity
DS1961_17_D_07	Up	3.04	peptinogen	Paralichthys olivaceus	BAC87742	3.00E-77	291	GO:0004194	pepsin A activity
DS1961_04_B_03	Up	4.65	pepsinogen A form IIa	Pseudopleuronectes americanus	AAE04984	e-105	384	GO:0004194	pepsin A activity
DS1961_15_G_05	Up	4.65	pepsinogen A form IIIa	Pseudopleuronectes americanus	AAE05203	3.00E-08	331	GO:0004194	pepsin A activity
DS1961_01_B_03	Up	3.41	pepsinogen C (progastricain)	Salvelinus fontinalis	AAE05046	e-107	390	GO:0004194	pepsin A activity
DS1961_66_H_07	Up	2.66	peptidylprolyl isomerase A (cyclophilin)	Danio rerio	AAE091263	2.00E-74	282	GO:0003755	peptidyl-prolyl cis-trans isomerase activity
DS1961_69_F_05	Up	2.77	peptidylprolyl isomerase A (cyclophilin)	Danio rerio	AAE091263	1.00E-61	239	GO:0003755	peptidyl-prolyl cis-trans isomerase activity
DS1961_74_E_05	Up	3.67	PKG(phosphoglycerate kinase 1) protein	Danio rerio	AAE65888	9.00E-84	313	GO:0006096	glycolysis
DS1961_55_F_04	Up	4.06	phosphoglucose isomerase-2	Plecoglossus altivelis	BAF91566	e-120	435	GO:0006096	glycolysis
DS1961_70_B_12	Up	3.43	phosphoglycerate kinase 1	Danio rerio	AAE65888	1.00E-35	137	GO:0006096	glycolysis
DS1961_22_A_04	Up	3.56	phosphoglycerate kinase 1	Danio rerio	AAE65888	5.00E-83	310	GO:0006096	glycolysis
DS1961_21_C_09	Up	2.22	sarcoendoplasmic reticulum calcium ATPase	Makaira nigricans	AAE808097	1.00E-83	313	GO:0006937	regulation of muscle contraction
DS1961_27_A_06	Up	2.45	sarcoendoplasmic reticulum calcium ATPase	Silurus asotus	AAE90494	2.00E-79	297	GO:0006937	regulation of muscle contraction
DS1961_27_A_01	Up	2.56	sarcoendoplasmic reticulum calcium ATPase	Silurus asotus	AAE90494	2.00E-77	279	GO:0006937	regulation of muscle contraction
DS1961_23_G_11	Up	2.71	sarcoendoplasmic reticulum calcium ATPase	Silurus asotus	AAE90494	6.00E-87	323	GO:0006937	regulation of muscle contraction
DS1961_62_B_06	Up	3.04	sarcoendoplasmic reticulum calcium ATPase 1A	Makaira nigricans	AAE808097	3.00E-81	305	GO:0006937	regulation of muscle contraction
DS1961_84_G_07	Up	2.26	sarcoendoplasmic reticulum calcium ATPase 1A	Makaira nigricans	AAE808097	3.00E-83	311	GO:0006937	regulation of muscle contraction
DS1961_70_C_05	Up	2.43	sarcoendoplasmic reticulum calcium ATPase 1A	Makaira nigricans	AAE808097	6.00E-85	317	GO:0006937	regulation of muscle contraction
DS1961_75_H_06	Up	2.86	selenoprotein P, Ia	Danio rerio	NP_840002	1.00E-53	213	GO:0001807	selenium metabolism
DS1961_69_G_06	Up	3.28	simple type II keratin K8b (32)	Oncorhynchus mykiss	CAE63300	3.00E-74	281	GO:0005882	intermediate filament
DS1961_77_B_06	Up	4.14	SFRAC1 secreted protein, acidic, rich in cysteine	Danio rerio	AAE01213	2.00E-31	139	GO:0000816	calcium ion transport
DS1961_32_E_04	Up	2.19	titin a	Danio rerio	AAE94500	3.00E-08	320	GO:0031432	titin binding
DS1961_36_G_11	Up	2.16	titin a	AAE94500	1.00E-69	333	GO:0031432	titin binding	
DS1961_07_D_03	Up	2.80	titin a	Danio rerio	AAE94500	e-125	451	GO:0031432	titin binding
DS1961_30_D_05	Up	1.59	transforming growth factor, beta-induced	Danio rerio	NP_878282	3.00E-21	105	GO:0000803	growth factor activity
DS1961_67_A_06	Up	2.21	transmembrane protein 30A	Danio rerio	NP_957194	6.00E-81	303	GO:0005267	potassium channel activity
DS1961_24_A_09	Up	2.20	tripartite motif-containing 45	Xenopus tropicalis	NP_001011026	3.00E-27	125	GO:0046872	metal ion binding
DS1961_34_B_11	Down	4.54	APEX nuclease (apurinic/apyrimidinic endonuclease) 2	Xenopus tropicalis	NP_001006804	6.00E-25	118	GO:0006261	DNA repair
DS1961_54_A_03	Down	1.69	calictonin receptor-like receptor	Oncorhynchus gibelus	CAB48406	5.00E-56	221	GO:0004948	calictonin receptor activity
DS1961_53_D_10	Down	1.58	calcium binding protein 39	Danio rerio	NP_998666	1.00E-76	290	GO:0019855	calcium channel inhibitor activity
DS1961_56_B_04	Down	2.01	cofilin 2 (muscle)	Danio rerio	NP_991243	5.00E-04	314	GO:0003779	actin binding
DS1961_59_A_01	Down	2.35	cofilin 2 (muscle)	Danio rerio	NP_991243	3.00E-83	311	GO:0003779	actin binding
DS1									

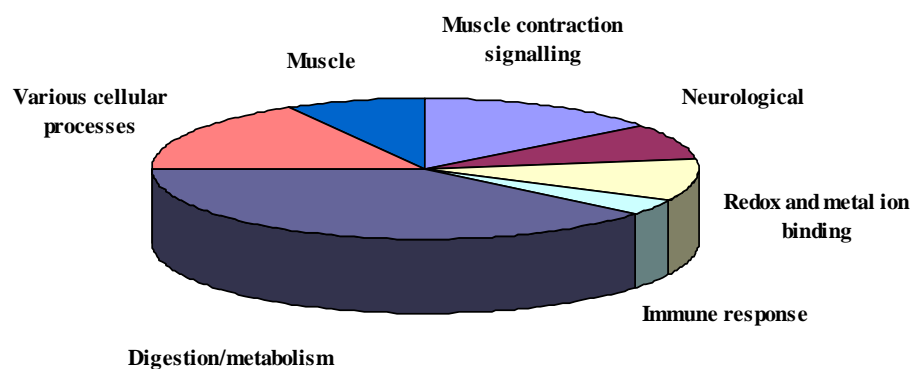


Figure 7-2. Functional classification of responding genes from 176 ESTs responding to copper (50 $\mu\text{g/L}$).

A subset of genes involved in redox and metal ion binding proteins were significantly affected during the 7-d exposure. Copper is an essential nutrient; an important part of many enzymes, normally found bound to proteins. At accumulated concentrations they may become free as highly reactive hydroxyl radicals. Oxidative damage by copper has been reported to cause abnormal Cu metabolism and neurodegenerative changes. Hemopexin was up-regulated by copper. Hemopexin induces the transcriptional activation of heme-oxygenase, are known to respond to nerve injury and may play a role in neurodegenerative disorders (Ferreira et al (1999). Gamma2-synuclein, a protein found primarily in the peripheral nervous system and implicated in neurodegenerative diseases (Surguchov et al., 2001) displayed differential expression. Corticotropin (lipotropin A precursor), a polypeptide hormone and neurotransmitter involved in the stress response was up-regulated and a glycine neurotransmitter transporter was down-regulated by copper.

Muscular activity in the delta smelt was affected by copper. Cardiac muscle actin was up-regulated in copper-exposed fish, as were myozenin; an α -actinin- and γ -filamin-binding Z line protein expressed predominantly in skeletal muscle, and sarcoendoplasmic ATPase; involved in the regulation of muscle contraction, alpha-tubulin, responsible for the formation of microtubules, was also up-regulated. In addition, muscle creatine kinase (up-regulated) is specifically bound to sarcoendoplasmic reticulum and can support calcium and uptake and regulate ATP/ADP ratios (Rossi et al., 1990), thus is directly involved in muscle contraction. Titin (also known as connectin) is an important protein also involved in muscle contraction, was up-regulated along with cofilin, an actin-binding factor required for the reorganization of actin

filaments.

Further genes involved in muscular activity and responding to copper exposure include those involved in calcium ion binding and potassium channel activity. Calcitonin receptor activity was up-regulated. Calcitonin is a hormone involved in bone mineral metabolism protecting the skeleton from calcium loss, and is also concerned with vitamin D regulation. Osteonectin (secreted protein acidic and rich in cysteine - SPARC) is a calcium-binding glycoprotein secreted by osteoblasts during bone formation, and was significantly up-regulated by copper in the delta smelt. Osteonectin is also a source of copper-binding peptides that are known to accumulate at sites of tissue repair (Lane et al., 1994). Elevated osteonectin expression has been reported to occur in a number of malignant tumors, and has been linked with inhibition of cancer cell metastasis (Koblinski et al., 2005) and has also been correlated with chronic pancreatitis (Bloomston et al., 2007). Interestingly, a gene encoding for a pancreatic protein with two somatomedin B domains was also up-regulated.

A number of digestive genes encoding for proteins involved in glycolysis, cholesterol efflux, lipid transport, chymotrypsin activity, proteolysis and other forms of digestion and metabolism were also seen to be affected by copper. Chitinase, a digestive enzyme that breaks down chitin was found to be up-regulated and is probably associated with food digestion (artemia exoskeleton in this test).

Lastly, immune responses were also seen to be affected. Down-regulated were tetraspanins; known to modulate the immune system and tumor necrosis factors (TNFs) involved in the regulation of immune cells and systemic inflammation. Changes in expression of these genes have been implicated in a variety of diseases. C1q complex genes, involved in immunoglobulin peptide fixation were up-regulated by copper exposure.

In summary, the overall responses to copper exposure in the delta smelt appear to be on neuro-muscular activity, respiration and metabolism as hypothesized. The immune system was also affected, and elevated expression of osteonectin may indicate tissue damage caused by excess copper. Confirmation tests are still required to verify the measured expression differences in greater detail. Real-time quantitative PCR will be undertaken to further investigate these responses.

Biomarker development, future work. Copper is the first of a suite of reference toxicants that are currently being assessed with the developed microarrays. From the responding genes, molecular biomarkers will be selected to quantitatively measure specific and general stress responses in the delta smelt to monitor the effect of water samples from the Sacramento San Joaquin watersheds and estuary upon their overall health. Chitinase and chymotrypsin for example, could give an indication of feeding activity and food digestion, whilst neurological impairments could be assessed using gamma synuclein and muscle activity by creatine kinase. Tetraspanin and TNFs would be an indicator of affected immune responses.

Real-time Quantitative PCR. A suite of real-time TaqMan PCR systems will be designed for selected ESTs responding significantly to copper exposure. For each target gene, two primers and an internal, fluorescent-labeled TaqMan probe (5' end, reporter dye FAM (6-carboxyfluorescein), 3' end, quencher dye TAMRA (6-carboxytetramethylrhodamine)) will be

designed using Primer Express software (Applied Biosystems, Foster City, USA). Relative gene expression (Livak and Schmittgen, 2001) or sequential normalization of target genes (Heckmann et al., 2006) will be used if no house-keeping genes are designated.

Genes for quantitative PCR currently selected from copper exposure:

- Gamma2-synuclein
- Hemopexin
- Creatine kinase
- myozenin
- Corticotropin
- Osteonectin
- Chitinase
- Tetraspanin
- Tumor Necrosis Factor (TNF)
- Cardiac muscle α -actin

We expect to assess these and genes from future microarray assessments in order to develop informative molecular biomarkers of stress and exposure in the delta smelt. We intend to carry out behavioral tests along with measurements of growth and survival for selected stressors.

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8. Quality Assurance/Quality Control

Quality Assurance/Quality Control (QA/QC) measures were included in this project to assess the reliability of the data collected. UCD ATL conducts approximately 10% of samples for QA/QC determinations. In 2006-2007, 9.6% of samples collected were slated for QA/QC (68 QA/QC samples were initiated in toxicity testing out of 710 total samples collected). These QA/QC procedures include positive control tests (i.e., reference toxicant tests), and QC samples such as field duplicates, bottle blanks and trip blanks. The components of these QA/QC measures are outlined below.

8.1 Reference Toxicant Tests

Positive control reference toxicant (RT) tests with *H. azteca* using NaCl as the toxicant were performed once a month to ascertain whether organism response fell within the acceptable range as dictated by US EPA. Each RT test consists of a dilution series made up for five different concentrations of the toxicant and a control. A 20-month running mean control chart is continuously updated with the results of these RT test endpoints. Acceptable range for US EPA is within the 95% confidence interval of a running mean. If the LC₅₀ or EC₂₅ falls out of the 95% confidence interval, test organism sensitivity is considered atypical and results of tests conducted during that month are considered suspect. Statistically speaking, one data point out of 20 will fall out of range by chance alone.

There were two months where *H. azteca* did not perform typically within the 95% confidence interval: February and June 2007 reference toxicant EC₂₅ values in weight exceeded the upper limit of the range. These outliers were instances in which an organism in the highest toxicant concentration survived, providing weight data where there previously was none, and normal EC₂₅ distribution was not obtained. Anomalous organism survival in higher RT toxicant concentrations for these months has not readily been explained, but the results indicate that *H. azteca* obtained for testing in the months of February and June could be less sensitive to potential contaminant(s) in ambient samples.

It is unlikely that test results in February and June, 2007 were affected by potentially less sensitive organisms, for survival LC₅₀ RT data consistently fell within the EPA RT range, and there were no statistically significant differences in organism survival among ambient samples and appropriate controls in tests conducted in the aforementioned months. Moreover, organisms utilized in toxicity tests conducted in February and June were sensitive enough to exhibit statistically significant differences in weight among ambient samples and the appropriate controls, and between PBO and non-manipulated ambient samples. However, it is understood that changes in organism sensitivity to a particular constituent such as NaCl may not necessarily affect an organism's sensitivity to other toxicant(s) that may be present in ambient samples utilized in toxicity testing. Therefore, February and June toxicity test data are considered reliable.

8.2 Field Duplicates

Field duplicate samples were collected to assess laboratory precision. A field duplicate sample is a second sample collected in a separate container(s), immediately after the primary test sample. Field duplicates are tested concurrently with its primary sample and the results are evaluated to determine precision of field and laboratory staff. Field duplicates were selected from *H. azteca*-specific sampling sites because QA/QC comparisons were not included in the developmental fish species bioassays. Field duplicate samples are in agreement when the primary sample and its duplicate are either statistically similar or statistically different from the control.

Twenty-one samples were collected as field duplicates in 2006; 18 samples were collected as field duplicates in 2007. In all instances, field duplicate samples were in agreement with their primary samples. Precision was determined by calculating the relative percent difference (RPD) between field duplicates and their primary samples in sample measurements. RPD is calculated by using the following equation:

$$RPD = \left(\frac{[2 * |Dup1 - Dup2|]}{[Dup1 + Dup2]} \right) * 100$$

Individual and average RPDs have been calculated for field duplicate samples collected in 2006 and 2007. Although this project does not fall under the Surface Water Ambient Monitoring Program (SWAMP), UCD ATL uses SWAMP QC guidelines in order to be comparable to other laboratories in California. SWAMP guidelines have a RPD limit of $\leq 35\%$ between duplicates. Field duplicate samples sharing equivalent results are listed in Table F1, and RPDs are listed in Tables F2-F10 (Appendix F).

8.3 Bottle Blanks

Bottle blank samples were included to evaluate potential incidental contamination due to the sampling container. Bottle blanks are analyte-free water samples that are transferred to a clean sample container that is prepared in the laboratory. For this project, bottle blanks were comprised of de-ionized water amended with dry salts to EPA moderately hard reconstituted specifications (DIEPAMHR). A bottle blank sample is in agreement when it is statistically similar to the control.

Six bottle blank samples were tested in 2006; 10 bottle blank samples were tested in 2007. With the exception of a bottle blank sample tested September 6, 2006; all bottle blanks shared equivalent results with the appropriate control. The bottle blank sample that was prepared in September, 2006 was not triple-rinsed prior to being filled with control water, and negatively affected the *H. azteca* weight endpoint. This was due to technician error. All laboratory staff were notified of the importance of triple-rinsing sample containers prior to use. Bottle blanks sharing equivalent results are outlined in Table F1 (Appendix F).

8.4 Trip Blanks

Trip blank samples were included to evaluate potential incidental contamination that can occur during field sampling and sample processing. A trip blank is an analyte-free water sample that is transferred into a clean sample container that is prepared in the laboratory, brought out into the field, and treated like any other collected sample throughout the course of the trip. For this project, trip blanks were comprised of DIEPAMHR. A trip blank sample is in agreement when it is statistically similar to the control.

Three trip blank samples were tested in 2006; 10 trip blanks were tested in 2007. All trip blank samples shared equivalent results with the appropriate control. Trip blanks sharing equivalent results are outlined in Table F1 (Appendix F).

8.5 Test Acceptability Criteria

Test acceptability criteria for *H. azteca* toxicity tests require 80% control survival. All *H. azteca* toxicity tests conducted in 2006 met all test acceptability criteria. All but two *H. azteca* toxicity tests conducted in 2007 met all test acceptability criteria. Tests in which control mortality exceeded 20% occurred with samples collected January 30/31, and April 11, 2007. In both cases the samples were re-initiated in secondary toxicity tests in which all test acceptability criteria were met. These data were considered reliable.

Test acceptability criteria for *M. saxatilis* and *H. transpacificus* require 80% control survival. These control limits were designated at the beginning of the project and were modeled after EPA chronic fish toxicity tests. After conducting two years of developmental toxicity testing with these species, it has been determined that these fish species are extremely sensitive at the ages utilized at UCD ATL and 80% control survival is not an attainable control limit. Therefore, only data in which control survival is less than 50% was rejected. All other data were considered reliable.

8.6 Deviations

Fourteen deviations occurred throughout the duration of the 2006-2007 POD project. Six deviations took place in 2006; 8 deviations occurred during 2007. The most frequent deviation were protocol deviations (4/14 or 29%), missed chemistry measurements (5/14 or 36%), high sample receiving temperatures (3/14 or 21%), and exceeded test initiation holding time (2/14 or 14%). Corrective actions were initiated whenever possible.

It is unlikely that these deviations had any negative impact on the data. Protocol deviations typically consisted of a reduced number of replicates; however there were

enough replicates initiated to achieve the statistical power needed to make comparisons between ambient treatments and appropriate controls. Missed chemistry measurements did not have an impact on the data, as organisms in those toxicity tests performed normally. High sample receiving temperatures had little to no effect on test data. While warm temperatures increase the chances of sample toxicant degradation, sample temperatures were close to the EPA criterion of 0-6 °C, and samples were placed in cold storage in the dark immediately upon receipt to negate any further degradation. Exceeded test initiation holding times were due to initial screening toxicity tests not meeting test acceptability criteria. In such cases, the samples were reinitiated in secondary toxicity tests, which exceeded test initiation holding time. This extended holding time may have resulted in loss of toxicant(s) due to sample degradation. However, samples were kept in the dark between 0-6 °C to minimize such degradation.

8.7 Completeness

Completeness is a measure of the data obtained compared to the amount of data expected in a project. The toxicity data acquisition phase of a project is considered complete when all sites specified in a contract have been visited the number of times designated in a contract, the number of samples designated in a contract has been collected, and the number of toxicity tests designated in the contract has been successfully completed. UCD ATL strives for a minimum of 90% completeness.

Over the course of 2006-2007, 100 *H. azteca* initial screening toxicity tests were conducted. Of those 100 tests, 98 passed all test acceptability criteria. The two tests which exhibited unacceptable control mortality were re-initiated and those retests met all test acceptability criteria. Therefore, 100% completeness was obtained for *H. azteca* toxicity testing.

As there are no standardized toxicity tests or completeness criteria established for *M. saxatilis* and *H. transpacificus* at this time, the completeness criterion cannot be determined for these species. Additional logistical factors make it difficult to determine completeness for these species. Large volumes of water are needed to initiate toxicity testing – up to 35 gallons of water are needed per sample. Such large volumes of water are difficult to obtain a second time if a test fails to meet test acceptability criteria, as samples are collected by boat through an external agency, which requires additional coordination. Manpower, boat availability and water storage can be problematic. Moreover, organisms are obtained through a commercial source. Due to the limited number of organisms available for testing (with *H. transpacificus* especially, as it is considered an endangered species), obtaining additional organisms to repeat a test is difficult. Additionally, UCD ATL is limited to the particular hatchery batch culture of organisms available for testing. As the commercial batch culture increases in age, it becomes nearly impossible to repeat a test if organisms utilized in that test were younger than the organisms available in the commercial batch. These species' sensitivity, combined with the aforementioned factors, make it nearly impossible to achieve a 90%

completeness criterion. Such logistical considerations should be taken into account in future project planning in order to maintain acceptable QA/QC criteria.

Appendix A
Hyalella azteca
Toxicity Tests

Table A1-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 5/28/07 examining the toxicity of piperonyl butoxide (PBO).

Treatment	Survival (%) ¹		Weight (mg/individual) ¹	
	mean	se	mean	se
DIEPAMHR	90	7.1	0.033	0.003
DIEPAMHR + 5 ppb PBO	90	7.1	0.040	0.006
DIEPAMHR + 10 ppb PBO	100	0.0	0.034	0.002
DIEPAMHR + 15 ppb PBO	100	0.0	0.044	0.005
DIEPAMHR + 20 ppb PBO	100	0.0	0.037	0.003
DIEPAMHR + 25 ppb PBO	98	2.5	0.039	0.005
DIEPAMHR + 50 ppb PBO	98	2.5	0.025	0.004
DIEPAMHR + 100 ppb PBO	98	2.5	0.021	0.001

Weight PMSD = 41.4%

Weight NOEC = 100 ppb

Weight EC25 = 42.4 ppb

1. Highlighted areas indicate a significant reduction in survival or weight compared to the DIEPAMHR control.

Table A2-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 01/13/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 01/12/06.

Treatment	Survival (%) ¹				
	Unmanipulated		100 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
Lab Control (DIEPAMHR)	96	4.2	100	0.0	NS
Low EC Control (Dilute DIEPAMHR)	100	0.0	100	0.0	NS
Old River, western arm at railroad bridge (902)	100	0.0	96	4.2	NS
Old River at mouth of Holland Cut (915)	96	4.2	100	0.0	NS
San Joaquin River between Hog and Turner Cuts (910)	100	0.0	100	0.0	NS
Sacramento River at tip of Grand Island (711) ³	100	0.0	100	0.0	NS
Sacramento R. across from Sherman Lake (704) ³	100	0.0	91	5.3	NS
Montezuma Slough at Nurse Slough (609)	100	0.0	96	4.2	NS
Suisun Bay, East of middle point (504) ³	100	0.0	100	0.0	NS
Suisun Bay off Chipps Island (508) ³	100	0.0	96	4.2	NS
Middle of Broad Slough, West end (804) ³	100	0.0	100	0.0	NS

	MSD	PMSD
One-way ANOVA	13.3	13.9
Two-way ANOVA	13.9	14.5

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		100 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
Lab Control (DIEPAMHR)	0.069	0.006	0.043	0.004	NS
Low EC Control (Dilute DIEPAMHR)	0.067	0.007	0.055	0.007	NS
Old River, western arm at railroad bridge (902)	0.118	0.009	0.107	0.014	NS
Old River at mouth of Holland Cut (915)	0.125	0.006	0.124	0.014	NS
San Joaquin River between Hog and Turner Cuts (910)	0.128	0.011	0.137	0.014	NS
Sacramento River at tip of Grand Island (711) ³	0.106	0.005	0.102	0.009	NS
Sacramento R. across from Sherman Lake (704) ³	0.119	0.008	0.100	0.023	NS
Montezuma Slough at Nurse Slough (609)	0.137	0.008	0.107	0.004	NS
Suisun Bay, East of middle point (504) ³	0.109	0.005	0.119	0.014	NS
Suisun Bay off Chipps Island (508) ³	0.118	0.016	0.101	0.011	NS
Middle of Broad Slough, West end (804) ³	0.109	0.008	0.098	0.011	NS

	MSD	PMSD
One-way ANOVA	0.041	59.6
Two-way ANOVA	0.054	77.8

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. This high conductivity sample was compared to the Low EC control @ 125 uS/cm

Table A2-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 1/12/06.

Treatment	Field Chemistry				Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)		
Old River, western arm at railroad bridge (902)	405	10.7	6.3	11.3	0.14	0.000
Old River at mouth of Holland Cut (915)	324	9.8	6.2	10.4	0.38	0.000
San Joaquin River between Hog and Turner Cuts (910)	249	10.4	6.6	11.1	0.27	0.000
Sacramento River at tip of Grand Island (711)	125	10.2	6.6	11.4	0.12	0.000
Sacramento R. across from Sherman Lake (704)	154	10.5	7.1	10.5	0.10	0.000
Montezuma Slough at Nurse Slough (609)	856	11.3	6.1	8.6	0.19	0.000
Suisun Bay, East of middle point (504)	186	10.9	6.3	9.9	0.14	0.000
Suisun Bay off Chipps Island (508)	157	10.3	6.5	10.3	0.09	0.000
Middle of Broad Slough, West end (804)	186	10.9	6.6	9.6	0.16	0.000

Table A2-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 1/13/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 1/12/06.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (µS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	392	21.3	23.9	6.5	8.7	7.47	8.10	92	58	-
Low EC Control	175	23.1	24.1	6.3	8.6	7.29	7.90	92	58	-
Old River, western arm at railroad bridge (902)	412	20.8	24.2	7.3	8.6	7.60	7.96	94	58	0.004
Old River at mouth of Holland Cut (915)	348	20.6	24.2	6.6	10.0	7.49	8.32	72	51	0.013
San Joaquin River between Hog and Turner Cuts (910)	237	20.9	24.3	6.8	11.2	7.42	8.91	56	46	0.010
Sacramento River at tip of Grand Island (711)	124	21.2	23.8	6.5	11.3	7.41	8.92	52	48	0.005
Sacramento R. across from Sherman Lake (704)	168	21.5	24.0	6.6	12.1	7.50	8.98	60	61	0.005
Montezuma Slough at Nurse Slough (609)	901	21.8	24.2	7.6	11.6	7.48	8.78	128	71	0.006
Suisun Bay, East of middle point (504)	205	21.8	24.0	6.9	12.6	7.48	9.08	56	55	0.006
Suisun Bay off Chipps Island (508)	163	23.0	23.7	6.7	11.9	7.50	8.92	56	60	0.003
Middle of Broad Slough, West end (804)	204	22.8	23.6	6.5	11.3	7.46	8.62	56	50	0.005
DIEPAMHR + 100 ppb PBO	399	21.7	24.7	6.0	8.3	7.44	8.05	-	-	-
Low EC Control + 100 ppb PBO	182	23.4	24.8	6.0	8.5	7.40	8.01	-	-	-
Old River, western arm at railroad bridge (902) + 100 ppb PBO	414	22.7	24.5	6.6	10.4	7.50	8.37	-	-	-
Old River at mouth of Holland Cut (915) + 100 ppb PBO	347	22.7	24.7	6.8	12.6	7.50	9.04	-	-	-
San Joaquin River between Hog and Turner Cuts (910) + 100 ppb PBO	239	23.3	24.6	6.7	12.8	7.53	9.19	-	-	-
Sacramento River at tip of Grand Island (711) + 100 ppb PBO	128	21.5	24.7	5.8	12.1	7.44	9.13	-	-	-
Sacramento R. across from Sherman Lake (704) + 100 ppb PBO	175	21.5	24.8	6.2	13.1	7.51	9.18	-	-	-
Montezuma Slough at Nurse Slough (609) + 100 ppb PBO	932	21.7	24.6	7.3	12.5	7.59	8.88	-	-	-
Suisun Bay, East of middle point (504) + 100 ppb PBO	215	22.0	24.5	6.3	13.1	7.53	9.17	-	-	-
Suisun Bay off Chipps Island (508) + 100 ppb PBO	167.3	22.8	24.7	6.4	14.4	7.59	9.50	-	-	-
Middle of Broad Slough, West end (804) + 100 ppb PBO	219	22.6	24.6	6.2	12.7	7.45	9.10	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A3-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 01/25/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 1/24/06.

Treatment	Survival (%) ¹	
	Unmanipulated	
	mean	se
Lab Control (DIEPAMHR)	100	0.0
Suisun Bay off Chipps Island (508)	100	0.0
Sacramento R. across from Sherman Lake (704)	96	4.0
Sacramento River at tip of Grand Island (711)	98	2.2
Middle of Broad Slough, West end (804)	96	4.0
Old River, western arm at railroad bridge (902)	92	5.8
San Joaquin River between Hog and Turner Cuts (910)	98	2.0
Old River at mouth of Holland Cut (915)	100	0.0

	MSD	PMSD
One-way ANOVA	- ²	- ²

Treatment	Weight (mg/Surviving individual) ¹	
	Unmanipulated	
	mean	se
Lab Control (DIEPAMHR)	0.066	0.005
Suisun Bay off Chipps Island (508)	0.106	0.004
Sacramento R. across from Sherman Lake (704)	0.080	0.003
Sacramento River at tip of Grand Island (711)	0.097	0.009
Middle of Broad Slough, West end (804)	0.114	0.010
Old River, western arm at railroad bridge (902)	0.107	0.005
San Joaquin River between Hog and Turner Cuts (910)	0.101	0.008
Old River at mouth of Holland Cut (915)	0.101	0.004

	MSD	PMSD
One-way ANOVA	- ²	- ²

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard statistical protocols.
2. Survival and weight were compared to the control using Kruskal-Wallis tests, and calculations of MSDs were not possible.

Table A3-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 01/24/06.

Treatment	Field Chemistry				Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)		
Suisun Bay off Chipps Island (508)	168.4	9.7	6.6	12.3	0.09	0.000
Sacramento R. across from Sherman Lake (704)	147.8	9.5	6.6	12.0	0.09	0.000
Sacramento River at tip of Grand Island (711)	135.0	9.7	7.1	10.3	0.09	0.000
Middle of Broad Slough, West end (804)	192.2	9.7	6.9	18.3	0.10	0.000
Old River, western arm at railroad bridge (902)	268.5	10.3	7.3	8.7	0.12	0.000
San Joaquin River between Hog and Turner Cuts (910)	215.4	10.5	6.8	8.3	0.42	0.000
Old River at mouth of Holland Cut (915)	351.4	10.2	6.8	8.8	0.12	0.000

Table A3-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 01/25/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 1/24/06.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
Lab Control (DIEPAMHR)	352.9	22.0	24.8	7.8	8.3	7.89	7.98	104	58	-
Suisun Bay off Chipps Island (508)	146.0	22.4	24.4	7.9	11.7	7.74	8.92	56	54	0.003
Sacramento R. across from Sherman Lake (704)	158.2	22.2	24.3	8.1	11.5	7.93	8.90	64	56	0.004
Sacramento River at tip of Grand Island (711)	153.6	22.2	24.3	8.0	10.8	7.80	8.72	44	44	0.003
Middle of Broad Slough, West end (804)	203.2	22.4	24.2	8.0	10.5	7.77	8.65	60	46	0.003
Old River, western arm at railroad bridge (902)	297.7	22.4	24.3	7.9	9.3	7.58	8.16	92	54	0.002
San Joaquin River between Hog and Turner Cuts (910)	320.8	22.0	24.1	8.0	10.7	7.55	8.76	56	46	0.007
Old River at mouth of Holland Cut (915)	354.3	23.3	24.2	7.6	9.1	7.50	8.22	72	50	0.002

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A4-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 01/26/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 01/25/06.

Treatment	Survival (%) ¹				
	Unmanipulated		100 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	99	1.1	99	1.0	NS
High EC Control @ 14.24 mS/cm	96	2.8	97	2.8	NS
Montezuma Slough at Nurse Slough (609)	100	0.0	100	0.0	NS
San Pablo Bay at Rodeo Flats (323)	94	2.4	51	20.1	S (54%)
Suisun Bay, East of middle point (504)	100	0.0	100	0.0	NS
Grizzly Bay at Dolphin (602)	100	0.0	100	0.0	NS
Carquinez Strait, West of Benicia army dock (405)	100	0.0	100	0.0	NS
Napa River at Riverside Blvd. terminus (340)	98	2.0	96	2.4	NS

	MSD	PMSD
One-way ANOVA ³	-	-
Two-way ANOVA	22.5	22.8

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		100 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.060	0.004	0.076	0.004	NS
High EC Control @ 14.24 mS/cm	0.070	0.005	0.061	0.004	NS
Montezuma Slough at Nurse Slough (609)	0.132	0.011	0.131	0.008	NS
San Pablo Bay at Rodeo Flats (323)	0.072	0.006	0.065	0.023	NS
Suisun Bay, East of middle point (504)	0.134	0.014	0.142	0.013	NS
Grizzly Bay at Dolphin (602)	0.164	0.009	0.162	0.015	NS
Carquinez Strait, West of Benicia army dock (405)	0.113	0.006	0.092	0.012	NS
Napa River at Riverside Blvd. terminus (340)	0.123	0.010	0.108	0.013	NS

	MSD	PMSD
One-way ANOVA	0.027	45.6
Two-way ANOVA	0.059	97.3

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. Survival in the unmanipulated samples was compared to the control using Wilcoxon Rank-Sum tests, and calculations of MSDs were not possible.

Table A4-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 01/25/06.

Treatment	Field Chemistry				Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)		
Montezuma Slough at Nurse Slough (609)	1068.0	9.9	7.70	9.2	0.13	0.001
San Pablo Bay at Rodeo Flats (323)	13830.0	10.7	7.60	9.9	0.09	0.001
Suisun Bay, East of middle point (504)	264.3	10.1	7.90	10.8	0.11	0.000
Grizzly Bay at Dolphin (602)	442.5	10.2	7.60	10.7	0.11	0.002
Carquinez Strait, West of Benicia army dock (405)	11190.0	10.6	7.50	10.0	0.14	0.001
Napa River at Riverside Blvd. terminus (340)	10270.0	10.7	7.70	9.8	0.10	0.001

Table A4-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 01/26/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 1/25/06.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	399	18.4	25.5	7.4	8.3	7.64	7.97	102	58	0.000
High EC Control @ 14.24 mS/cm	14240	18.2	24.2	7.1	8.3	7.43	7.71	-	-	-
Montezuma Slough at Nurse Slough (609)	991	19.2	25.4	6.4	11.8	7.67	8.67	156	75	0.006
San Pablo Bay at Rodeo Flats (323)	13000	18.6	25.4	7.4	12.3	7.69	8.23	1506	75	0.002
Suisun Bay, East of middle point (504)	345	18.5	25.4	7.1	11.5	7.68	8.87	68	60	0.003
Grizzly Bay at Dolphin (602)	461	18.7	25.3	7.5	11.2	7.70	8.71	92	66	0.005
Carquinez Strait, West of Benicia army dock (405)	12785	18.8	25.1	7.6	11.0	7.54	8.25	1272	74	0.001
Napa River ar Riverside Blvd. terminus (340)	10160	19.8	25.2	7.9	9.5	7.63	7.91	1092	78	0.002
DIEPAMHR + 100 ppb PBO	456	17.9	25.3	6.9	8.1	7.64	7.98	-	-	-
High EC Control @ 14.24 mS/cm + 100 ppb PBO	14240	18.2	24.2	7.1	8.3	7.43	7.71	-	-	-
Montezuma Slough at Nurse Slough (609) + 100 ppb PBO	1120	20.3	25.0	6.8	9.9	7.83	8.34	-	-	-
San Pablo Bay at Rodeo Flats (323) + 100 ppb PBO	14560	18.2	25.0	7.8	9.0	7.68	7.91	-	-	-
Suisun Bay, East of middle point (504) + 100 ppb PBO	365	17.9	25.1	6.0	10.2	7.77	8.85	-	-	-
Grizzly Bay at Dolphin (602) + 100 ppb PBO	402	17.6	25.1	7.1	9.0	7.85	8.00	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 100 ppb PBO	12285	17.7	25.1	7.3	8.8	7.47	7.53	-	-	-
Napa River ar Riverside Blvd. terminus (340) + 100 ppb PBO	10195	17.7	25.1	7.2	8.2	7.46	7.66	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A5-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 02/08/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 02/07/06.

Treatment	Survival (%) ¹				
	Unmanipulated		100 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
Lab Control (DIEPAMHR)	94	2.4	98	2.0	NS
Low EC Control (Dilute DIEPAMHR)	92	3.8	95	3.1	NS
Sacramento Deep Water Channel (Light 55)	100	0.0	100	0.0	NS
Sacramento R. across from Sherman Lake (704)	100	0.0	100	0.0	NS
Sacramento River at tip of Grand Island (711) ³	98	2.0	100	0.0	NS
Middle of Broad Slough, West end (804)	100	0.0	98	2.0	NS
San Joaquin River, West of Oulton Point (812)	100	0.0	98	2.0	NS
Old River, western arm at railroad bridge (902)	98	2.0	100	0.0	NS
San Joaquin River between Hog and Turner Cuts (910)	100	0.0	96	2.4	NS
Old River at mouth of Holland Cut (915)	100	0.0	98	2.0	NS

	MSD	PMSD
One-way ANOVA ⁴	-	-
Two-way ANOVA	10.3	11.0

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		100 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
Lab Control (DIEPAMHR)	0.033	0.006	0.025	0.004	NS
Low EC Control (Dilute DIEPAMHR)	0.023	0.005	0.015	0.002	NS
Sacramento Deep Water Channel (Light 55)	0.042	0.002	0.062	0.004	NS
Sacramento R. across from Sherman Lake (704)	0.038	0.002	0.044	0.004	NS
Sacramento River at tip of Grand Island (711) ³	0.048	0.006	0.043	0.007	NS
Middle of Broad Slough, West end (804)	0.041	0.001	0.048	0.007	NS
San Joaquin River, West of Oulton Point (812)	0.033	0.006	0.053	0.006	NS
Old River, western arm at railroad bridge (902)	0.031	0.005	0.048	0.003	NS
San Joaquin River between Hog and Turner Cuts (910)	0.036	0.004	0.054	0.004	NS
Old River at mouth of Holland Cut (915)	0.049	0.003	0.062	0.006	NS

	MSD	PMSD
One-way ANOVA	0.016	49.2
Two-way ANOVA	0.028	114.0

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. This low conductivity sample was compared to the low conductivity control.

4. Survival in the unmanipulated samples was compared to the control using Steel's Many-One Rank Test, and calculation of an MSD was not possible.

Table A5-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 02/07/06.

Treatment	Field Chemistry				Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)		
Sacramento Deep Water Channel (Light 55)	96	10.6	7.67	13.9	0.12	0.005
Sacramento R. across from Sherman Lake (704)	131	12.5	7.41	13.5	0.10	0.003
Sacramento River at tip of Grand Island (711)	114	10.0	7.50	13.9	0.13	0.003
Middle of Broad Slough, West end (804)	261	10.5	7.95	12.6	0.11	0.003
San Joaquin River, West of Oulton Point (812)	94	10.6	7.59	13.6	0.10	0.003
Old River, western arm at railroad bridge (902)	248	10.7	7.58	10.7	0.07	0.002
San Joaquin River between Hog and Turner Cuts (910)	291	11.5	7.40	12.3	0.23	0.007
Old River at mouth of Holland Cut (915)	296	11.1	7.48	13.3	0.07	0.001

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Table A5-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 02/08/06 of samples collected by the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 02/07/06.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
Lab Control (DIEPAMHR)	366.2	16.5	24.6	6.0	8.2	7.6	7.84	108	62	-
Low EC Lab Control (Dilute DIEPAMHR)	121.9	17.1	24.5	6.1	8.3	7.31	7.51	24	18	-
Sacramento Deep Water Channel (Light 55)	183.1	17.5	24.6	6.6	11.0	7.65	8.34	68	70	0.005
Sacramento R. across from Sherman Lake (704)	141.6	17.3	24.6	6.4	10.2	7.52	8.31	52	52	0.003
Sacramento River at tip of Grand Island (711)	124.7	17.2	24.6	6.2	10.7	7.48	8.32	44	48	0.003
Middle of Broad Slough, West end (804)	189.0	17.6	24.6	6.1	10.1	7.46	8.08	50	52	0.003
San Joaquin River, West of Oulton Point (812)	182.8	17.8	24.7	5.8	9.7	7.48	8.03	52	52	0.003
Old River, western arm at railroad bridge (902)	255.1	17.5	24.7	5.9	9.1	7.48	7.93	72	52	0.002
San Joaquin River between Hog and Turner Cuts (910)	303.6	18.7	24.7	6.1	10.2	7.54	8.22	72	54	0.006
Old River at mouth of Holland Cut (915)	307.5	21.5	24.6	6.2	10.9	7.54	8.70	72	56	0.001
Lab Control (DIEPAMHR) + 100 ppb PBO	389.5	23.9	24.5	6.2	8.3	7.55	7.87	-	-	-
Low EC Lab Control (Dilute DIEPAMHR) + 100 ppb PBO	127.0	18.8	24.5	7.0	8.1	7.35	7.61	-	-	-
Sacramento Deep Water Channel (Light 55) + 100 ppb PBO	187.6	17.9	24.7	7.5	10.2	7.72	8.40	-	-	-
Sacramento R. across from Sherman Lake (704) + 100 ppb PBO	143.3	18.9	24.6	7.2	10.7	7.62	8.48	-	-	-
Sacramento River at tip of Grand Island (711) + 100 ppb PBO	128.6	19	24.6	6.8	10.1	7.58	8.15	-	-	-
Middle of Broad Slough, West end (804) + 100 ppb PBO	193.1	19.3	24.5	6.5	8.9	7.54	7.96	-	-	-
San Joaquin River, West of Oulton Point (812) + 100 ppb PBO	181.2	18.5	24.5	6.7	9.3	7.56	8.00	-	-	-
Old River, western arm at railroad bridge (902) + 100 ppb PBO	254.3	18.6	24.5	6.4	9.1	7.52	7.91	-	-	-
San Joaquin River between Hog and Turner Cuts (910) + 100 ppb PBO	301.5	19	24.6	6.7	9.3	7.57	7.97	-	-	-
Old River at mouth of Holland Cut (915) + 100 ppb PBO	295.0	19	24.6	6.9	9.8	7.61	8.16	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A6-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 02/09/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 02/08/06.

Treatment	Survival (%) ¹				
	Unmanipulated		100 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	100	0.0	96	2.3	NS
High EC Control	100	0.0	90	4.5	NS
San Pablo Bay at Rodeo Flats (323)	98	2.0	93	6.7	NS
Napa River ar Riverside Blvd. terminus (340)	100	0.0	96	2.4	NS
Carquinez Strait, West of Benicia army dock (405)	100	0.0	100	0.0	NS
Suisun Bay, East of middle point (504)	98	2.0	96	2.4	NS
Suisun Bay off Chipps Island (508)	100	0.0	92	3.5	NS
Grizzly Bay at Dolphin (602)	100	0.0	100	0.0	NS
Montezuma Slough at Nurse Slough (609)	100	0.0	100	0.0	NS

	MSD	PMSD
One-way ANOVA ³	-	-
Two-way ANOVA	14.0	14.0

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		100 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.053	0.004	0.027	0.006	NS
High EC Control	0.034	0.004	0.028	0.005	NS
San Pablo Bay at Rodeo Flats (323)	0.058	0.003	0.043	0.006	NS
Napa River ar Riverside Blvd. terminus (340)	0.053	0.006	0.060	0.003	NS
Carquinez Strait, West of Benicia army dock (405)	0.073	0.005	0.086	0.004	NS
Suisun Bay, East of middle point (504)	0.073	0.008	0.079	0.009	NS
Suisun Bay off Chipps Island (508)	0.079	0.006	0.086	0.010	NS
Grizzly Bay at Dolphin (602)	0.090	0.006	0.087	0.003	NS
Montezuma Slough at Nurse Slough (609)	0.084	0.008	0.089	0.003	NS

	MSD	PMSD
One-way ANOVA ³	-	-
Two-way ANOVA	0.035	65.3

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. Survival and weight in the unmanipulated samples were compared to the control using Steel's Many-One Rank Test, and calculations of MSDs were not possible.

Table A6-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 02/08/06.

Treatment	Field Chemistry				Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)		
San Pablo Bay at Rodeo Flats (323)	10812	11.6	7.63	10.0	0.10	0.002
Napa River ar Riverside Blvd. terminus (340)	9310	11.4	7.57	9.9	0.15	0.002
Carquinez Strait, West of Benicia army dock (405)	1118	10.9	7.58	10.5	0.11	0.002
Suisun Bay, East of middle point (504)	266	10.9	7.59	10.4	0.10	0.002
Suisun Bay off Chipps Island (508)	157	10.9	7.50	10.5	0.11	0.002
Grizzly Bay at Dolphin (602)	321	10.9	7.65	10.4	0.17	0.004
Montezuma Slough at Nurse Slough (609)	1023	10.9	7.35	9.1	0.17	0.002

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Table A6-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 02/09/06 of samples collected by the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 02/08/06.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
Lab Control (DIEPAMHR)	386	22.1	25.0	7.4	8.8	7.68	8.16	108	62	-
High EC Lab Control (DIEPAMHR + Salt Water)	11575	21.9	25.2	7.3	9.2	7.59	7.83	-	-	-
San Pablo Bay at Rodeo Flats (323)	11095	21.8	25.3	7.7	9.2	7.62	7.74	1240	80	0.002
Napa River ar Riverside Blvd. terminus (340)	9905	21.6	25.3	7.8	9.2	7.54	7.76	976	80	0.002
Carquinez Strait, West of Benicia army dock (405)	1418	21.9	25.3	7.2	9.0	7.72	7.93	152	65	0.003
Suisun Bay, East of middle point (504)	268	22.2	25.2	7.2	9.1	7.78	7.99	64	60	0.003
Suisun Bay off Chipps Island (508)	195	21.6	25.2	6.8	9.4	7.67	7.89	56	58	0.003
Grizzly Bay at Dolphin (602)	365	21.7	25.4	7.4	9.1	7.57	7.90	80	65	0.003
Montezuma Slough at Nurse Slough (609)	650	21.7	24.8	7.0	8.7	7.41	7.72	156	76	0.002
Lab Control (DIEPAMHR) + 100 ppb PBO	365	21.0	24.9	7.7	8.8	7.82	8.11	-	-	-
High EC Lab Control (DIEPAMHR + Salt Water) + 100 ppb PBO	10995	21.5	24.9	6.9	9.0	7.38	7.88	-	-	-
San Pablo Bay at Rodeo Flats (323) + 100 ppb PBO	10820	21.8	24.9	8.3	13.5	7.6	8.65	-	-	-
Napa River ar Riverside Blvd. terminus (340) + 100 ppb PBO	9220	21.3	25.0	7.8	11.1	7.55	8.18	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 100 ppb PBO	1531	21.6	25.0	7.7	11.3	7.77	8.64	-	-	-
Suisun Bay, East of middle point (504) + 100 ppb PBO	293	21.9	24.9	6.8	10.9	7.8	8.72	-	-	-
Suisun Bay off Chipps Island (508) + 100 ppb PBO	183	21.9	25.0	7.7	11.8	7.62	8.99	-	-	-
Grizzly Bay at Dolphin (602) + 100 ppb PBO	342	22.1	25.0	7.2	12.3	7.53	8.93	-	-	-
Montezuma Slough at Nurse Slough (609) + 100 ppb PBO	1033	21.7	21.7	7.3	11.4	7.4	8.67	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A7-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 02/22/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 02/21/06.

Treatment	Survival (%) ¹				
	Unmanipulated		100 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
Lab Control (DIEPAMHR)	100	0.0	100	0.0	NS
Sacramento Deep Water Channel (Light 55)	100	0.0	98	2.0	NS
Sacramento River at tip of Grand Island (711)	94	4.0	98	2.0	NS
Middle of Broad Slough, West end (804)	100	0.0	98	2.0	NS
San Joaquin River, West of Oulton Point (812)	98	2.0	100	0.0	NS
Old River, western arm at railroad bridge (902)	100	0.0	100	0.0	NS
San Joaquin River between Hog and Turner Cuts (910)	100	0.0	100	0.0	NS
Old River at mouth of Holland Cut (915)	98	2.0	98	2.0	NS

	MSD	PMSD
One-way ANOVA ³	-	-
Two-way ANOVA	8.5	8.5

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		100 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
Lab Control (DIEPAMHR)	0.065	0.008	0.048	0.006	NS
Sacramento Deep Water Channel (Light 55)	0.124	0.007	0.097	0.001	NS
Sacramento River at tip of Grand Island (711)	0.083	0.001	0.072	0.004	NS
Middle of Broad Slough, West end (804)	0.088	0.006	0.087	0.003	NS
San Joaquin River, West of Oulton Point (812)	0.101	0.009	0.069	0.008	NS
Old River, western arm at railroad bridge (902)	0.110	0.013	0.069	0.004	NS
San Joaquin River between Hog and Turner Cuts (910)	0.114	0.011	0.095	0.018	NS
Old River at mouth of Holland Cut (915)	0.111	0.013	0.090	0.009	NS

	MSD	PMSD
One-way ANOVA	0.047	71.7
Two-way ANOVA	0.052	80.1

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. Survival in the unmanipulated samples was compared to the control using a Kruskal-Wallis test, and calculation of an MSD was not possible.

Table A7-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 02/21/06.

Treatment	Field Chemistry				Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)		
Sacramento Deep Water Channel (Light 55)	321	11.3	7.78	11.3	0.03	0.000
Sacramento River at tip of Grand Island (711)	141	9.8	7.53	11.5	0.12	0.001
Middle of Broad Slough, West end (804)	164	8.4	7.97	10.2	0.00	0.000
San Joaquin River, West of Oulton Point (812)	156	10.8	7.42	10.2	0.02	0.000
Old River, western arm at railroad bridge (902)	189	10.3	7.65	10.3	0.00	0.000
San Joaquin River between Hog and Turner Cuts (910)	332	11.1	7.36	9.8	0.30	0.001
Old River at mouth of Holland Cut (915)	210	10.6	7.44	10.0	0.00	0.000
Field Dup.: Old River at mouth of Holland Cut (915)	210	10.6	7.44	10.0	0.00	0.000

Table A7-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 02/22/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 02/21/06.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
Lab Control (DIEPAMHR)	366	21.8	23.3	6.9	8.6	7.64	8.08	116	58	0.000
Sacramento Deep Water Channel (Light 55)	263	22.2	23.4	7.2	10.1	7.82	7.98	128	138	0.001
Sacramento River at tip of Grand Island (711)	192	21.8	23.5	6.7	10.2	7.62	7.94	64	66	0.005
Middle of Broad Slough, West end (804)	191	21.4	23.5	7.0	9.3	7.53	7.82	60	58	0.000
San Joaquin River, West of Oulton Point (812)	204	21.7	23.3	6.9	9.2	7.50	7.54	64	58	0.000
Old River, western arm at railroad bridge (902)	222	21.5	23.3	6.9	9.2	7.45	7.64	64	56	0.000
San Joaquin River between Hog and Turner Cuts (910)	371	21.5	23.3	7.4	9.8	7.34	7.53	84	59	0.003
Old River at mouth of Holland Cut (915)	263	21.5	23.3	7.7	9.1	7.54	7.56	68	57	0.000
Field Dup.: Old River at mouth of Holland Cut (915)	248	21.4	23.2	7.3	9.1	7.52	7.66	68	55	0.000
Lab Control (DIEPAMHR) + 100 ppb PBO	378	19.7	22.5	7.2	9.3	7.50	7.99	-	-	-
Sacramento Deep Water Channel (Light 55) + 100 ppb PBO	360	19.3	23.0	7.3	8.6	7.89	8.18	-	-	-
Sacramento River at tip of Grand Island (711) + 100 ppb PBO	186	19.3	23.2	6.2	8.8	7.58	8.09	-	-	-
Middle of Broad Slough, West end (804) + 100 ppb PBO	203	19.4	23.2	6.7	8.7	7.51	8.07	-	-	-
San Joaquin River, West of Oulton Point (812) + 100 ppb PBO	201	19.4	23.2	5.9	8.8	7.43	7.93	-	-	-
Old River, western arm at railroad bridge (902) + 100 ppb PBO	217	20.6	23.1	6.6	9.4	7.52	7.71	-	-	-
San Joaquin River between Hog and Turner Cuts (910) + 100 ppb PBO	377	20.0	23.2	7.0	9.2	7.53	7.90	-	-	-
Old River at mouth of Holland Cut (915) + 100 ppb PBO	259	19.4	23.2	6.7	9.2	7.51	7.68	-	-	-
Field Dup.: Old River at mouth of Holland Cut (915) + 100 ppb PBO	269	18.7	22.9	6.8	8.7	7.52	7.79	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A8-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 02/23/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 02/22/06.

Treatment	Survival (%) ¹				
	Unmanipulated		100 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	100	0.0	100	0.0	NS
High EC Control @ 17.30 mS/cm	100	0.0	-	-	-
San Pablo Bat at Rodeo Flats (323) ³	96	4.4	100	0.0	NS
Napa River ar Riverside Blvd. terminus (340) ³	100	0.0	100	0.0	NS
Carquinez Strait, West of Benicia army dock (405) ³	100	0.0	98	1.8	NS
Suisun Bay, East of middle point (504)	100	0.0	100	0.0	NS
Suisun Bay off Chipps Island (508)	98	2.0	100	0.0	NS
Grizzly Bay at Dolphin (602)	100	0.0	100	0.0	NS
Montezuma Slough at Nurse Slough (609)	100	0.0	100	0.0	NS
Sacramento R. across from Sherman Lake (704)	96	2.3	100	0.0	NS

	MSD	PMSD
One-way ANOVA ⁴	-	-
Two-way ANOVA	6.7	6.7

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		100 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.087	0.010	0.041	0.006	NS
High EC Control @ 17.30 mS/cm	0.056	0.004	-	-	-
San Pablo Bat at Rodeo Flats (323) ³	0.083	0.016	0.043	0.013	NS
Napa River ar Riverside Blvd. terminus (340) ³	0.099	0.011	0.076	0.006	NS
Carquinez Strait, West of Benicia army dock (405) ³	0.110	0.009	0.097	0.011	NS
Suisun Bay, East of middle point (504)	0.109	0.005	0.111	0.011	NS
Suisun Bay off Chipps Island (508)	0.144	0.009	0.117	0.003	NS
Grizzly Bay at Dolphin (602)	0.113	0.012	0.140	0.006	NS
Montezuma Slough at Nurse Slough (609)	0.123	0.016	0.167	0.018	NS
Sacramento R. across from Sherman Lake (704)	0.098	0.003	0.111	0.004	NS

	MSD	PMSD
One-way ANOVA	0.047	53.9
Two-way ANOVA	0.053	61.3

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. These high conductivity treatments were compared to the high EC control.

4. Survival in the unmanipulated samples was compared to the control using a Kruskal-Wallis test, and calculation of an MSD was not possible.

Table A8-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 02/22/06.

Treatment	Field Chemistry				Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)		
San Pablo Bay at Rodeo Flats (323)	16311	10.8	7.83	10.0	0.06	0.001
Napa River at Riverside Blvd. terminus (340)	15151	10.7	7.72	9.4	0.09	0.001
Carquinez Strait, West of Benicia army dock (405)	12074	10.8	7.73	9.7	0.10	0.001
Suisun Bay, East of middle point (504)	2370	11.3	7.78	10.5	0.11	0.001
Suisun Bay off Chipps Island (508)	764	11.3	7.68	10.4	0.10	0.001
Grizzly Bay at Dolphin (602)	4843	10.4	7.66	10.1	0.11	0.001
Montezuma Slough at Nurse Slough (609)	800	10.5	7.54	9.0	0.18	0.001
Sacramento R. across from Sherman Lake (704)	175	9.9	7.92	10.5	0.15	0.002

Table A8-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 02/23/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 02/22/06.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	375	23.2	23.6	6.1	8.4	7.51	8.25	116	58	-
High EC Control @ 17.30 mS/cm	17930	22.6	24.0	7.3	8.3	7.55	8.01	-	-	-
San Pablo Bat at Rodeo Flats (323)	17110	22.6	24.0	7.0	8.4	7.54	7.91	1968	84	0.002
Napa River ar Riverside Blvd. terminus (340)	15735	22.8	24.2	7.6	8.4	7.60	7.85	1752	86	0.002
Carquinez Strait, West of Benicia army dock (405)	13450	22.4	23.9	7.9	8.2	7.63	7.88	1496	82	0.003
Suisun Bay, East of middle point (504)	2629	22.5	23.8	7.1	8.4	7.68	8.16	276	70	0.006
Suisun Bay off Chipps Island (508)	888	22.3	24.1	7.1	8.4	7.85	8.29	124	72	0.008
Grizzly Bay at Dolphin (602)	4811	22.1	24.1	7.4	8.4	7.56	7.94	520	78	0.004
Montezuma Slough at Nurse Slough (609)	950	22.0	23.9	6.8	8.3	7.81	8.23	136	80	0.013
Sacramento R. across from Sherman Lake (704)	235	22.6	23.0	7.1	8.6	7.93	8.26	72	72	0.012
DIEPAMHR + 100 ppb PBO	364	22.2	23.3	6.6	8.7	7.63	8.25	-	-	-
High EC Control @ 17.30mS/cm + 100 ppb PBO	16765	21.6	23.3	7.6	8.4	7.44	8.02	-	-	-
San Pablo Bat at Rodeo Flats (323) + 100 ppb PBO	16520	21.5	23.2	7.6	8.8	7.49	7.87	-	-	-
Napa River ar Riverside Blvd. terminus (340) + 100 ppb PBO	14965	20.8	23.2	7.8	8.4	7.54	7.89	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 100 ppb PBO	12615	20.8	23.2	7.4	9.0	7.55	8.04	-	-	-
Suisun Bay, East of middle point (504) + 100 ppb PBO	2702	21.3	23.4	6.7	8.7	7.58	8.41	-	-	-
Suisun Bay off Chipps Island (508) + 100 ppb PBO	854	20.3	23.1	6.8	9.8	7.76	8.36	-	-	-
Grizzly Bay at Dolphin (602) + 100 ppb PBO	4455	22.2	23.1	6.6	9.6	7.47	8.03	-	-	-
Montezuma Slough at Nurse Slough (609) + 100 ppb PBO	981	22.0	23.3	6.7	9.1	7.77	8.20	-	-	-
Sacramento R. across from Sherman Lake (704) + 100 ppb PBO	225	22.1	23.3	6.8	9.8	7.90	8.37	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A9-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 3/08/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 3/07/06.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	100	0.0	90	5.8	S (90%)
Low EC Control (Dilute DIEPAMHR)	98	2.0	-	-	NA
Sacramento R. Deep Water Channel, Light 55	100	0.0	100	0.0	NS
Sacramento River at tip of Grand Island (711) ³	100	0.0	100	0.0	NS
Broad Slough, West End (804) ³	100	0.0	96	3.7	NS
SJR @ West of Oulton Point (812) ³	100	0.0	100	0.0	NS
Old River @ Holland Cut (902)	100	0.0	100	0.0	NS
SJR @ Hong and Turner Cut (910)	100	0.0	100	0.0	NS
Old River @ RR Bridge, West (915)	100	0.0	97	3.0	NS
Field Duplicate: Broad Slough, West End (804) ³	100	0.0	97	3.0	NS

	MSD	PMSD
One-way ANOVA ⁴	-	-
Two-way ANOVA	7.8	7.8

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.099	0.015	0.064	0.006	NS
Low EC Control (Dilute DIEPAMHR)	0.046	0.006	-	-	NA
Sacramento R. Deep Water Channel, Light 55	0.139	0.017	0.093	0.009	NS
Sacramento River at tip of Grand Island (711) ³	0.091	0.004	0.083	0.012	NS
Broad Slough, West End (804) ³	0.127	0.008	0.107	0.010	NS
SJR @ West of Oulton Point (812) ³	0.112	0.016	0.118	0.018	NS
Old River @ Holland Cut (902)	0.111	0.008	0.108	0.007	NS
SJR @ Hong and Turner Cut (910)	0.141	0.010	0.114	0.012	NS
Old River @ RR Bridge, West (915)	0.107	0.013	0.112	0.009	NS
Field Duplicate: Broad Slough, West End (804) ³	0.123	0.011	0.104	0.009	NS

	MSD	PMSD
One-way ANOVA	0.055	55.2
Two-way ANOVA	0.064	64.7

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard statistical protocols.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. This low conductivity sample was compared to the Low EC control.

4. Survival in the unmanipulated samples was compared to the control using a Kruskal-Wallis test, and calculation of an MSD was not possible.

Table A9-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 03/07/2006.

Treatment	Field Chemistry				Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)		
Sacramento R. Deep Water Channel, Light 55	250	11.75	7.77	10.22	0.10	0.0012
Sacramento River at tip of Grand Island (711)	149	11.89	7.84	10.75	0.08	0.0011
Broad Slough, West End (804)	160	10.77	7.62	10.34	0.08	0.0006
SJR @ West of Oulton Point (812)	133	10.55	7.46	10.60	0.09	0.0005
Old River @ Holland Cut (902)	218	11.55	7.53	9.84	0.03	0.0002
San Joaquin River between Hog and Turner Cuts (910)	266	11.61	7.47	10.14	0.29	0.0017
Old River @ RR Bridge, West (915)	253	11.91	7.58	10.04	0.04	0.0003
Field Duplicate: Broad Slough, West End (804)	160	10.77	7.62	10.34	0.09	0.0007

Table A9-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 03/08/2006 of samples collected by the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 03/07/2006.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	381	21.5	24.3	6.3	8.0	7.57	8.09	104	58	-
Low EC Control (Dilute DIEPAMHR)	133	19.0	26.7	7.3	8.6	7.34	8.31	-	-	-
Sacramento R. Deep Water Channel, Light 55	206	21.2	24.3	7.4	13.8	7.75	9.09	412	72	0.0028
Sacramento River at tip of Grand Island (711)	4891	21.3	24.2	7.4	12.2	7.65	8.71	180	42	0.0022
Broad Slough, West End (804)	4835	21.3	24.5	7.1	10.8	7.57	8.34	80	52	0.0021
SJR @ West of Oulton Point (812)	4695	21.3	24.7	7.4	12.4	7.67	8.76	72	50	0.0027
Old River @ Holland Cut (902)	244	21.2	24.6	7.5	10.6	7.00	8.19	56	58	0.0002
San Joaquin River between Hog and Turner Cuts (910)	296	21.0	24.5	7.6	13.0	7.72	9.01	64	48	0.0096
Old River @ RR Bridge, West (915)	281	21.3	24.4	7.5	10.4	7.69	8.27	160	58	0.0011
Field Duplicate: Broad Slough, West End (804)	172	21.2	24.5	7.3	12.5	7.61	8.94	76	52	0.0019
DIEPAMHR + 100 ppb PBO	376	20.9	24.2	6.8	8.1	7.57	8.15	-	-	-
Low EC Control + 100 ppb PBO	218	20.9	24.2	7.6	9.1	7.80	8.04	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 100 ppb PBO	125	21.1	24.2	7.8	9.3	7.65	7.96	-	-	-
Sacramento River at tip of Grand Island (711) + 100 ppb PBO	186	20.6	24.3	7.4	8.4	7.66	7.94	-	-	-
Broad Slough, West End (804) + 100 ppb PBO	161	20.7	24.3	7.6	9.3	7.54	8.09	-	-	-
SJR @ West of Oulton Point (812) + 100 ppb PBO	245	19.7	23.9	7.5	8.3	7.71	8.00	-	-	-
Old River @ Holland Cut (902) + 100 ppb PBO	298	20.6	24.3	7.9	9.5	7.66	8.09	-	-	-
San Joaquin River between Hog and Turner Cuts (910) + 100 ppb PBO	285	19.5	24.0	7.9	8.9	7.72	7.95	-	-	-
Old River @ RR Bridge, West (915) + 100 ppb PBO	222	20.7	24.0	9.0	9.5	7.61	7.92	-	-	-
Field Duplicate: Broad Slough, West End (804) + 100 ppb PBO	133	19.0	26.7	7.3	8.6	7.34	8.31	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A10-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 03/08/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 03/09/06.

Treatment	Survival (%) ¹				
	Unmanipulated		100 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	98	1.8	100	0.0	NS
San Pablo Bat at Rodeo Flats (323)	100	0.0	100	0.0	NS
Napa River at Vallejo Seawall (340)	100	0.0	100	0.0	NS
Carquinez Strait, West of Benicia army dock (405)	100	0.0	100	0.0	NS
Suisun Bay, East of middle point (504)	98	2.0	100	0.0	NS
Suisun Bay off Chipps Island (508)	100	0.0	100	0.0	NS
Grizzly Bay at Dolphin (602)	100	0.0	100	0.0	NS
Montezuma Slough at Nurse Slough (609)	98	2.0	100	0.0	NS
Sacramento R. across from Sherman Lake (704)	100	0.0	100	0.0	NS
Bottle Blank	100	0.0	100	0.0	NS

	MSD	PMSD
One-way ANOVA ³	-	-
Two-way ANOVA	5.4	5.5

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		100 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.102	0.007	0.074	0.003	NS
San Pablo Bat at Rodeo Flats (323)	0.128	0.005	0.114	0.007	NS
Napa River at Vallejo Seawall (340)	0.143	0.006	0.132	0.006	NS
Carquinez Strait, West of Benicia army dock (405)	0.153	0.009	0.137	0.017	NS
Suisun Bay, East of middle point (504)	0.123	0.008	0.119	0.008	NS
Suisun Bay off Chipps Island (508)	0.126	0.013	0.117	0.012	NS
Grizzly Bay at Dolphin (602)	0.152	0.004	0.123	0.010	NS
Montezuma Slough at Nurse Slough (609)	0.154	0.007	0.135	0.005	NS
Sacramento R. across from Sherman Lake (704)	0.134	0.004	0.111	0.006	NS
Bottle Blank	0.094	0.008	-	-	-

	MSD	PMSD
One-way ANOVA	0.036	35.0
Two-way ANOVA	0.043	42.7

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Unmanipulated samples were analyzed using USEPA standard statistical protocols. Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.
3. Survival in the unmanipulated samples was compared to the control using a Kruskal-Wallis test, and calculation of an MSD was not possible.

Table A10-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 03/08/06.

Treatment	Field Chemistry				Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)		
San Pablo Bay at Rodeo Flats (323)	3670	11.2	7.67	10.4	0.14	0.001
Napa River at Vallejo Seawall (340)	1368	11.0	7.61	10.1	0.15	0.001
Carquinez Strait, West of Benicia army dock (405)	324	10.6	7.70	10.4	0.09	0.001
Suisun Bay, East of middle point (504)	143	10.6	7.66	10.6	0.10	0.001
Suisun Bay off Chipps Island (508)	168	10.4	7.76	10.5	0.09	0.001
Grizzly Bay at Dolphin (602)	154	10.7	7.70	10.6	0.09	0.001
Montezuma Slough at Nurse Slough (609)	837	11.5	7.30	9.8	0.21	0.001
Sacramento R. across from Sherman Lake (704)	178	10.8	7.79	10.5	0.05	0.001

Table A10-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 03/08/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 03/09/06.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
Lab Control (DIEPAMHR)	379.7	20.3	25.1	6.7	8.3	7.65	8.05	104	58	-
San Pablo Bat at Rodeo Flats (323)	3767.5	20.1	25.8	7.9	8.5	7.66	7.86	412	72	0.005
Napa River at Vallejo Seawall (340)	1334.5	20.4	25.7	7.6	8.2	7.65	7.84	180	64	0.005
Carquinez Strait, West of Benicia army dock (405)	427.3	20.7	25.7	7.6	8.2	7.67	7.90	80	64	0.004
Suisun Bay, East of middle point (504)	176.9	20.4	25.5	7.7	8.8	7.61	7.98	72	54	0.004
Suisun Bay off Chipps Island (508)	191.5	20.4	25.4	7.5	8.6	7.60	8.10	56	66	0.006
Grizzly Bay at Dolphin (602)	175.0	20.4	25.1	7.7	8.6	7.65	7.89	64	62	0.003
Montezuma Slough at Nurse Slough (609)	876.5	20.6	25.0	7.8	9.5	7.53	8.19	160	66	0.004
Sacramento R. across from Sherman Lake (704)	244.2	20.9	24.9	8.1	8.8	7.82	8.13	76	72	0.002
Bottle Blank	377.3	20.8	25.1	7.3	8.6	7.59	7.94	-	-	-
DIEPAMHR + 100 ppb PBO	359.3	20.8	23.7	6.7	8.9	7.60	8.12	-	-	-
San Pablo Bat at Rodeo Flats (323) + 100 ppb PBO	3514.0	20.0	23.9	8.0	10.9	7.53	8.47	-	-	-
Napa River at Vallejo Seawall (340) + 100 ppb PBO	1297.0	20.1	23.9	7.4	9.5	7.57	8.16	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 100 ppb PBO	412.2	20.8	24.0	7.2	10.1	7.64	8.45	-	-	-
Suisun Bay, East of middle point (504) + 100 ppb PBO	168.6	21.1	24.0	7.5	10.3	7.63	8.72	-	-	-
Suisun Bay off Chipps Island (508) + 100 ppb PBO	173.3	21.1	24.0	7.2	11.8	7.71	8.98	-	-	-
Grizzly Bay at Dolphin (602) + 100 ppb PBO	170.2	21.8	23.9	7.4	11.4	7.69	8.99	-	-	-
Montezuma Slough at Nurse Slough (609) + 100 ppb PBO	828.0	21.3	23.9	7.4	10.9	7.62	8.73	-	-	-
Sacramento R. across from Sherman Lake (704) + 100 ppb PBO	209.8	21.5	23.9	7.2	12.6	7.79	9.20	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A11-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 03/20/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 03/21/06.

Treatment	Survival (%) ¹				
	Unmanipulated		100 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	100	0.0	95	5.0	NS
Sacramento Deep Water Channel (Light 55)	100	0.0	100	0.0	NS
Sacramento R. across from Sherman Lake (704)	100	0.0	100	0.0	NS
Sacramento River at tip of Grand Island (711)	98	2.0	100	0.0	NS
Middle of Broad Slough, West end (804)	100	0.0	100	0.0	NS
San Joaquin River, West of Oulton Point (812)	100	0.0	100	0.0	NS
Old River, western arm at railroad bridge (902)	98	2.0	100	0.0	NS
San Joaquin River between Hog and Turner Cuts (910)	96	2.4	100	0.0	NS
Old River at mouth of Holland Cut (915)	98	2.0	100	0.0	NS
Field Dup: San Joaquin River between Hog and Turner Cuts (910)	100	0.0	100	0.0	NS

	MSD	PMSD
One-way ANOVA ³	-	-
Two-way ANOVA	9.1	9.1

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		100 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.055	0.007	0.042	0.006	NS
Sacramento Deep Water Channel (Light 55)	0.109	0.014	0.080	0.005	NS
Sacramento R. across from Sherman Lake (704)	0.116	0.010	0.104	0.018	NS
Sacramento River at tip of Grand Island (711)	0.098	0.011	0.083	0.030	NS
Middle of Broad Slough, West end (804)	0.109	0.014	0.105	0.014	NS
San Joaquin River, West of Oulton Point (812)	0.105	0.011	0.109	0.008	NS
Old River, western arm at railroad bridge (902)	0.083	0.022	0.109	0.009	NS
San Joaquin River between Hog and Turner Cuts (910)	0.123	0.010	0.085	0.001	NS
Old River at mouth of Holland Cut (915)	0.093	0.008	0.108	0.001	NS
Field Dup: San Joaquin River between Hog and Turner Cuts (910)	0.140	0.015	0.095	0.032	NS

	MSD	PMSD
One-way ANOVA	0.055	100.3
Two-way ANOVA	0.074	135.3

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard statistical protocols.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. Survival in the unmanipulated samples was compared to the control using a Kruskal-Wallis test, and calculation of an MSD was not possible.

Table A11-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 03/20/06.

Treatment	Field Chemistry				Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)		
Sacramento Deep Water Channel (Light 55)	416	11.4	8.10	10.4	0.07	0.002
Sacramento R. across from Sherman Lake (704)	207	10.8	8.04	10.7	0.07	0.001
Sacramento River at tip of Grand Island (711)	122	10.2	7.69	10.9	0.09	0.001
Middle of Broad Slough, West end (804)	175	10.7	7.94	10.7	0.07	0.001
San Joaquin River, West of Oulton Point (812)	158	10.7	7.66	10.5	0.09	0.001
Old River, western arm at railroad bridge (902)	239	11.5	7.77	10.3	0.03	0.000
San Joaquin River between Hog and Turner Cuts (910)	223	11.5	7.57	10.1	0.23	0.002
Old River at mouth of Holland Cut (915)	319	11.9	7.80	10.5	0.02	0.000
Field Dup: San Joaquin River between Hog and Turner Cuts (910)	223	11.5	7.57	10.1	0.23	0.002

Table A11-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 03/20/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 03/21/06.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	387	21.5	24.9	6.6	8.0	7.73	8.13	108	60	-
Sacramento Deep Water Channel (Light 55)	463	21.6	24.9	6.7	8.7	7.96	8.25	132	112	0.006
Sacramento R. across from Sherman Lake (704)	235	21.4	24.9	6.8	9.6	7.91	8.52	88	88	0.007
Sacramento River at tip of Grand Island (711)	152	21.6	24.6	6.6	9.0	7.71	8.02	52	54	0.005
Middle of Broad Slough, West end (804)	201	21.6	24.7	6.9	8.4	7.74	8.03	60	56	0.004
San Joaquin River, West of Oulton Point (812)	196	21.3	24.7	6.6	8.4	7.68	7.98	60	60	0.004
Old River, western arm at railroad bridge (902)	286	21.6	24.7	6.5	8.7	7.65	8.04	68	54	0.002
San Joaquin River between Hog and Turner Cuts (910)	255	21.2	24.6	6.8	8.8	7.66	8.02	60	50	0.012
Old River at mouth of Holland Cut (915)	439	22.0	24.7	6.4	8.3	7.67	7.93	92	62	0.001
Field Dup: San Joaquin River between Hog and Turner Cuts (910)	250	21.5	24.8	6.8	8.6	7.65	8.04	72	48	0.012
DIEPAMHR + 100 ppb PBO	377	20.9	24.2	6.6	8.1	7.74	8.15	-	-	-
Sacramento Deep Water Channel (Light 55) + 100 ppb PBO	443	20.9	24.2	7.1	10.0	8.02	8.78	-	-	-
Sacramento R. across from Sherman Lake (704) + 100 ppb PBO	225	21.6	24.2	7.7	11.1	8.07	9.01	-	-	-
Sacramento River at tip of Grand Island (711) + 100 ppb PBO	141	22.1	24.3	7.5	11.1	7.86	9.11	-	-	-
Middle of Broad Slough, West end (804) + 100 ppb PBO	187	21.5	24.2	6.7	9.6	7.75	8.51	-	-	-
San Joaquin River, West of Oulton Point (812) + 100 ppb PBO	178	20.9	24.2	7.0	9.9	7.71	8.57	-	-	-
Old River, western arm at railroad bridge (902) + 100 ppb PBO	261	20.8	24.3	7.2	8.9	7.78	8.12	-	-	-
San Joaquin River between Hog and Turner Cuts (910) + 100 ppb PBO	244	20.7	24.2	7.7	9.8	7.78	8.54	-	-	-
Old River at mouth of Holland Cut (915) + 100 ppb PBO	350	20.9	24.2	7.1	9.0	7.73	8.29	-	-	-
Field Dup: San Joaquin R., Hog and Turner Cuts (910) + 100 ppb PBO	237	21.6	24.3	7.0	10.7	7.69	8.89	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A12-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 03/22/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 03/21/06.

Treatment	Survival (%) ¹				
	Unmanipulated		100 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	100	0.0	100	0.0	NS
High EC Control @ 8000 uS/cm	100	0.0	-	-	NA
San Pablo Bay at Rodeo Flats (323)	100	0.0	97	3.3	NS
Napa River at Riverside Blvd. terminus (340)	100	0.0	100	0.0	NS
Carquinez Strait, West of Benicia army dock (405)	100	0.0	100	0.0	NS
Suisun Bay, East of middle point (504)	100	0.0	100	0.0	NS
Suisun Bay off Chipps Island (508)	100	0.0	100	0.0	NS
Grizzly Bay at Dolphin (602)	100	0.0	100	0.0	NS
Montezuma Slough at Nurse Slough (609)	100	0.0	100	0.0	NS

	MSD	PMSD
One-way ANOVA ³	-	-
Two-way ANOVA	3.1	3.1

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		100 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
Lab Control (DIEPAMHR)	0.076	0.004	0.056	0.004	NS
High EC Control @ 8000 uS/cm	0.061	0.003	-	-	-
San Pablo Bay at Rodeo Flats (323)	0.120	0.009	0.084	0.008	NS
Napa River at Riverside Blvd. terminus (340)	0.132	0.006	0.103	0.005	NS
Carquinez Strait, West of Benicia army dock (405)	0.123	0.005	0.114	0.015	NS
Suisun Bay, East of middle point (504)	0.122	0.006	0.078	0.009	S (64%)
Suisun Bay off Chipps Island (508)	0.103	0.009	0.081	0.002	NS
Grizzly Bay at Dolphin (602)	0.124	0.009	0.107	0.003	NS
Montezuma Slough at Nurse Slough (609)	0.141	0.006	0.119	0.008	NS

	MSD	PMSD
One-way ANOVA	0.031	41.1
Two-way ANOVA	0.039	51.4

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard statistical protocols.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. Survival in the unmanipulated samples was compared to the control using a Kruskal-Wallis test, and calculation of an MSD was not possible.

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Table A12-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 03/21/06.

Treatment	Field Chemistry				Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)		
San Pablo Bay at Rodeo Flats (323)	8179	11.1	7.66	10.1	0.11	0.001
Napa River at Riverside Blvd. terminus (340)	2679	11.2	7.62	9.9	0.18	0.001
Carquinez Strait, West of Benicia army dock (405)	545	10.9	7.66	10.8	0.05	0.000
Suisun Bay, East of middle point (504)	235	10.8	7.89	10.6	0.06	0.001
Suisun Bay off Chipps Island (508)	138	10.4	7.64	10.8	0.05	0.000
Grizzly Bay at Dolphin (602)	158	10.4	7.84	10.8	0.05	0.001
Montezuma Slough at Nurse Slough (609)	237	10.5	7.70	10.3	0.09	0.001

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Table A12-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 03/22/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 03/21/06.

Treatment	Laboratory Chemistry						Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹	
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH				Max pH
DIEPAMHR	401	21	25.1	5.7	8.1	7.52	8.30	108	60	-
High EC Control @ 8000 uS/cm	8150	19.7	25.1	6.3	8.3	7.52	8.25	-	-	-
San Pablo Bay at Rodeo Flats (323)	2941	20.1	25.0	6.1	9.7	7.61	7.95	1232	84	0.001
Napa River ar Riverside Blvd. terminus (340)	697	20.4	25.1	6.4	11.4	7.63	7.69	400	108	0.005
Carquinez Strait, West of Benicia army dock (405)	221	20.2	25.2	7.1	10.3	7.76	7.85	132	68	0.001
Suisun Bay, East of middle point (504)	178	20.7	25.0	6.4	11.0	7.68	7.83	80	70	0.002
Suisun Bay off Chipps Island (508)	188	20.5	25.2	6.7	11.9	7.71	7.82	72	66	0.002
Grizzly Bay at Dolphin (602)	288	20.8	25.3	6.2	10.9	7.67	7.74	96	76	0.002
Montezuma Slough at Nurse Slough (609)	356	21.4	23.5	6.3	8.4	7.57	8.19	92	74	0.003
DIEPAMHR + 100 ppb PBO	10445	20.5	25.1	6.2	9.4	7.48	8.52	-	-	-
San Pablo Bay at Rodeo Flats (323) + 100 ppb PBO	10000	21.1	23.6	6.4	8.6	7.53	8.78	-	-	-
Napa River ar Riverside Blvd. terminus (340) + 100 ppb PBO	3142	21.5	23.7	6.2	8.5	7.61	9.01	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 100 ppb PBO	703	21.2	23.8	6.3	9.4	7.64	9.11	-	-	-
Suisun Bay, East of middle point (504) + 100 ppb PBO	215	21.7	23.7	4.4	8.4	7.46	8.89	-	-	-
Suisun Bay off Chipps Island (508) + 100 ppb PBO	177	21.6	23.8	6.4	9.3	7.65	8.51	-	-	-
Grizzly Bay at Dolphin (602) + 100 ppb PBO	199	22.1	23.8	6.4	10.4	7.67	8.57	-	-	-
Montezuma Slough at Nurse Slough (609) + 100 ppb PBO	285	21.7	23.8	6.2	9.2	7.66	8.12	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A13-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 4/5/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 4/03/06 - 4/04/06.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	94	4.0	62	31.2	NS
Low EC Control @ 100 uS/cm	96	2.5	-	-	NA
High EC Control @ 7000 uS/cm	100	0.0	-	-	NA
Sacramento Deep Water Channel (Light 55)	100	0.0	100	0.0	NS
Sacramento River at tip of Grand Island (711) ³	98	2.0	77	23.3	NS
Old River, western arm at railroad bridge (902)	88	12.5	97	2.8	NS
San Joaquin River between Hog and Turner Cuts (910)	100	0.0	100	0.0	NS
Old River at mouth of Holland Cut (915)	98	1.8	61	30.8	NS
San Pablo Bay at Rodeo Flats (323) ⁴	96	2.6	100	0.0	NS
Napa River at Vallejo Seawall (340)	98	2.0	100	0.0	NS
Carquinez Strait, West of Benicia army dock (405)	98	1.8	100	0.0	NS
Suisun Bay off Chipps Island (508)	100	0.0	97	3.0	NS
Montezuma Slough at Nurse Slough (609)	100	0.0	97	3.0	NS
San Joaquin River, West of Oulton Point (812)	62	21.3	100	0.0	NS
Field Dup.: Old River at mouth of Holland Cut (915)	87	13.3	81	19.0	NS
Field Dup.: Napa River at Vallejo Seawall (340)	98	2.0	100	0.0	NS

	MSD	PMSD
One-way ANOVA ⁵	-	-
Two-way ANOVA	40.9	43.5

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.060	0.013	0.050	0.026	NS
Low EC Control @ 100 uS/cm	0.061	0.010	-	-	NA
High EC Control @ 7000 uS/cm	0.060	0.003	-	-	NA
Sacramento Deep Water Channel (Light 55)	0.110	0.003	0.093	0.020	NS
Sacramento River at tip of Grand Island (711) ³	0.758	0.007	0.066	0.012	NS
Old River, western arm at railroad bridge (902)	0.101	0.019	0.086	0.011	NS
San Joaquin River between Hog and Turner Cuts (910)	0.118	0.008	0.073	0.003	NS
Old River at mouth of Holland Cut (915)	0.110	0.006	0.068	0.017	NS
San Pablo Bay at Rodeo Flats (323) ⁴	0.068	0.011	0.093	0.021	NS
Napa River at Vallejo Seawall (340)	0.119	0.008	0.093	0.009	NS
Carquinez Strait, West of Benicia army dock (405)	0.145	0.008	0.117	0.015	NS
Suisun Bay off Chipps Island (508)	0.095	0.010	0.091	0.011	NS
Montezuma Slough at Nurse Slough (609)	0.149	0.010	0.114	0.004	NS
San Joaquin River, West of Oulton Point (812)	0.086	0.006	0.100	0.003	NS
Field Dup.: Old River at mouth of Holland Cut (915)	0.107	0.011	0.097	0.014	NS
Field Dup.: Napa River at Vallejo Seawall (340)	0.127	0.006	0.080	0.012	NS

	MSD	PMSD
One-way ANOVA	0.047	79.5
Two-way ANOVA	0.061	102.2

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. This high conductivity sample was compared to the Low EC control @ 100 uS/cm.

4. This high conductivity sample was compared to the High EC control @ 7000 uS/cm.

5. Survival in the unmanipulated samples was compared to the control using a Kruskal-Wallis test, and calculation of an MSD was not possible.

Table A13-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 4/03/06 - 4/04/06.

Treatment	Field Chemistry				Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)		
Sacramento Deep Water Channel (Light 55)	178	11.8	7.82	10.5	0.04	0.001
Sacramento River at tip of Grand Island (711)	95	10.8	7.64	11.0	0.09	0.001
Old River, western arm at railroad bridge (902)	254	13.2	7.73	9.8	0.02	0.000
San Joaquin River between Hog and Turner Cuts (910)	157	13.1	7.57	9.9	0.23	0.002
Old River at mouth of Holland Cut (915)	200	12.9	7.56	9.7	0.07	0.001
San Pablo Bay at Rodeo Flats (323)	9520	12.1	7.56	9.4	0.11	0.001
Napa River at Vallejo Seawall (340)	838	12.2	7.59	9.1	0.15	0.001
Carquinez Strait, West of Benicia army dock (405)	647	12.0	7.58	10.5	0.09	0.001
Suisun Bay off Chipps Island (508)	155	11.6	7.55	10.2	0.08	0.001
Montezuma Slough at Nurse Slough (609)	607	11.9	7.27	10.1	0.13	0.000
San Joaquin River, West of Oulton Point (812)	221	12.2	7.09	10.3	0.10	0.000
Field Dup.: Old River at mouth of Holland Cut (915)	200	12.9	7.56	9.7	0.02	0.000
Field Dup.: Napa River at Vallejo Seawall (340)	838	12.2	7.59	9.1	0.18	0.001

Table A13-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 4/05/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 4/03/06 - 4/04/06.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	384	21.4	25.2	6.8	8.2	7.59	8.17	112	62	-
Low EC Control @ 100 uS/cm	149	23.3	25.1	6.7	8.5	7.35	7.94	-	-	-
High EC Control @ 7000 uS/cm	7945	21.5	22.4	7.7	9.2	7.43	7.86	-	-	-
Sacramento Deep Water Channel (Light 55)	243	20.4	25.5	7.3	8.9	7.67	8.07	80	80	0.002
Sacramento River at tip of Grand Island (711)	134	20.6	25.7	7.3	8.5	7.56	8.08	52	48	0.006
Old River, western arm at railroad bridge (902)	308	19.7	25.8	7.1	9.5	7.55	8.08	76	56	0.001
San Joaquin River between Hog and Turner Cuts (910)	203	19.9	25.7	7.2	8.2	7.52	8.05	60	54	0.014
Old River at mouth of Holland Cut (915)	289	19.9	25.3	7.1	8.0	7.59	8.23	60	48	0.006
San Pablo Bay at Rodeo Flats (323)	9190	20.2	25.4	7.4	8.4	7.48	7.82	1024	76	0.003
Napa River at Vallejo Seawall (340)	942	23.8	25.4	7.0	8.0	7.61	8.09	140	76	0.009
Carquinez Strait, West of Benicia army dock (405)	1103	22.8	25.4	7.4	8.0	7.57	7.99	152	68	0.004
Suisun Bay off Chipps Island (508)	204	22.1	25.4	7.3	8.1	7.64	8.15	68	56	0.006
Montezuma Slough at Nurse Slough (609)	627	22.2	24.8	7.2	8.5	7.72	8.14	128	78	0.008
San Joaquin River, West of Oulton Point (812)	249	21.4	25.0	7.1	8.8	7.53	8.06	56	50	0.006
Field Dup.: Old River at mouth of Holland Cut (915)	236	20.3	25.0	7.3	9.1	7.73	8.16	76	80	0.001
Field Dup.: Napa River at Vallejo Seawall (340)	1230	20.4	25.5	6.9	8.3	7.78	8.18	112	76	0.013
DIEPAMHR + 100 ppb PBO	370	20.4	24.2	7.4	8.4	7.71	8.19	-	-	-
Sacramento Deep Water Channel (Light 55) + 100 ppb PBO	229	19.5	24.2	7.5	8.4	7.85	8.12	-	-	-
Sacramento River at tip of Grand Island (711) + 100 ppb PBO	137	19.5	24.3	7.3	8.2	7.59	8.09	-	-	-
Old River, western arm at railroad bridge (902) + 100 ppb PBO	349	20.0	24.3	7.3	8.1	7.63	7.87	-	-	-
San Joaquin River between Hog and Turner Cuts (910) + 100 ppb PBO	201	20.1	24.3	7.3	8.0	7.54	8.05	-	-	-
Old River at mouth of Holland Cut (915) + 100 ppb PBO	236	20.3	24.2	7.2	8.1	7.54	8.04	-	-	-
San Pablo Bay at Rodeo Flats (323) + 100 ppb PBO	8780	20.5	24.2	7.5	8.1	7.45	7.95	-	-	-
Napa River at Vallejo Seawall (340) + 100 ppb PBO	904	21.0	24.2	7.4	8.1	7.51	8.11	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 100 ppb PBO	1092	21.8	24.2	7.5	8.1	7.64	8.08	-	-	-
Suisun Bay off Chipps Island (508) + 100 ppb PBO	198	21.6	24.2	7.7	8.2	7.67	8.05	-	-	-
Montezuma Slough at Nurse Slough (609) + 100 ppb PBO	663	22.6	24.2	7.5	7.9	7.70	8.20	-	-	-
San Joaquin River, West of Oulton Point (812) + 100 ppb PBO	249	22.6	24.3	6.8	8.1	7.45	8.08	-	-	-
Field Dup.: Old River at mouth of Holland Cut (915) + 100 ppb PBO	235	19.4	24.3	7.7	8.2	7.89	8.05	-	-	-
Field Dup.: Napa River at Vallejo Seawall (340) + 100 ppb PBO	1122	20.8	24.2	7.3	8.1	7.60	8.04	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A14-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 4/06/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 4/05/06.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	100	0.0	97	3.3	NS
Suisun Bay, East of middle point (504)	98	2.0	70	29.6	NS
Grizzly Bay at Dolphin (602)	100	0.0	100	0.0	NS
Sacramento R. across from Sherman Lake (704)	100	0.0	94	3.2	NS
Middle of Broad Slough, West end (804)	96	2.4	100	0.0	NS
Field Dup.: Grizzly Bay at Dolphin (602)	100	0.0	100	0.0	NS
Trip Blank	98	2.2	100	0.0	NS

	MSD	PMSD
One-way ANOVA ³	-	-
Two-way ANOVA	30.6	30.6

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.057	0.008	0.043	0.016	NS
Suisun Bay, East of middle point (504)	0.079	0.008	0.043	0.048	NS
Grizzly Bay at Dolphin (602)	0.095	0.008	0.079	0.010	NS
Sacramento R. across from Sherman Lake (704)	0.095	0.003	0.075	0.009	NS
Middle of Broad Slough, West end (804)	0.115	0.016	0.097	0.014	NS
Field Dup.: Grizzly Bay at Dolphin (602)	0.084	0.010	0.093	0.025	NS
Trip Blank	0.071	0.005	0.035	0.005	NS

	MSD	PMSD
One-way ANOVA	0.042	73.2
Two-way ANOVA	0.064	111.4

- Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$). Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
- NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.
- Survival in the unmanipulated samples was compared to the control using a Kruskal-Wallis test, and calculation of an MSD was not possible.

Table A14-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 4/05/06.

Treatment	Field Chemistry				Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)		
Suisun Bay, East of middle point (504)	161	11.6	7.66	10.4	0.08	0.001
Grizzly Bay at Dolphin (602)	183	11.7	7.74	10.3	0.06	0.001
Sacramento R. across from Sherman Lake (704)	179	11.3	7.78	10.4	0.07	0.001
Middle of Broad Slough, West end (804)	191	11.9	8.34	11.0	0.07	0.003
Field Dup.: Grizzly Bay at Dolphin (602)	183	11.7	7.74	10.3	0.04	0.000
Trip Blank	339	18.7	8.24	9.6	0.00	0.000

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Table A14-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 4/06/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 4/05/06.

Treatment	Laboratory Chemistry						Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹	
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH				Max pH
DIEPAMHR	347	20.9	25.0	7.0	8.1	7.70	8.05	112	62	-
Suisun Bay, East of middle point (504)	172	22.3	25.3	7.1	8.6	7.63	7.86	60	60	0.003
Grizzly Bay at Dolphin (602)	192	22.4	25.1	7.1	8.4	7.62	7.92	72	72	0.003
Sacramento R. across from Sherman Lake (704)	202	22.4	25.5	7.0	8.6	7.69	7.95	76	80	0.003
Middle of Broad Slough, West end (804)	175	22.3	25.5	7.0	7.9	7.55	7.88	60	50	0.003
Field Dup.: Grizzly Bay at Dolphin (602)	199	21.5	25.0	7.2	8.6	7.74	7.82	68	72	0.001
Trip Blank	338	22.7	25.5	6.9	8.2	7.64	7.93	104	60	0.000
DIEPAMHR + 100 ppb PBO	344	21.3	24.4	7.5	8.2	7.72	8.04	-	-	-
Suisun Bay, East of middle point (504) + 100 ppb PBO	165	22.4	24.6	7.1	8.7	7.69	7.84	-	-	-
Grizzly Bay at Dolphin (602) + 100 ppb PBO	201	22.8	24.6	7.2	8.5	7.78	8.02	-	-	-
Sacramento R. across from Sherman Lake (704) + 100 ppb PBO	203	22.6	24.7	6.8	9.2	7.78	7.80	-	-	-
Middle of Broad Slough, West end (804) + 100 ppb PBO	171	22.7	24.6	7.4	8.3	7.60	7.92	-	-	-
Field Dup.: Grizzly Bay at Dolphin (602) + 100 ppb PBO	200	22.5	24.6	7.2	8.7	7.77	7.85	-	-	-
Trip Blank + 100 ppb PBO	352	22.8	24.6	7.2	8.4	7.67	7.87	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A15-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 4/18/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 4/17/06.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	98	2.0	100	0.0	NS
Low EC Control @ 100 uS/cm	98	2.2	67	33.3	NS
Sacramento Deep Water Channel, Light 55	100	0.0	100	0.0	NS
Sacramento R. across from Sherman Lake (704)	100	0.0	100	0.0	NS
Sacramento River at tip of Grand Island (711) ³	100	0.0	93	3.3	NS
Middle of Broad Slough, West end (804)	92	8.0	78	22.2	NS
San Joaquin River, West of Oulton Point (812)	100	0.0	93	6.7	NS
Old River, western arm at railroad bridge (902)	100	0.0	100	0.0	NS
San Joaquin River between Hog and Turner Cuts (910)	100	0.0	67	33.3	NS
Old River at mouth of Holland Cut (915)	100	0.0	90	10.0	NS
Field Dup.: San Joaquin River between Hog and Turner Cuts (910)	100	0.0	-	-	NA

	MSD	PMSD
One-way ANOVA	13.9	14.2
Two-way ANOVA	25.2	25.7

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.085	0.011	0.056	0.001	NS
Low EC Control @ 100 uS/cm	0.047	0.006	0.037	0.008	NS
Sacramento Deep Water Channel, Light 55	0.102	0.011	0.115	0.007	NS
Field Dup.: San Joaquin River between Hog and Turner Cuts (910)	0.143	0.011	-	-	NA
Sacramento R. across from Sherman Lake (704)	0.109	0.007	0.069	0.012	NS
Sacramento River at tip of Grand Island (711) ³	0.088	0.014	0.056	0.014	NS
Middle of Broad Slough, West end (804)	0.099	0.023	0.085	0.012	NS
San Joaquin River, West of Oulton Point (812)	0.108	0.019	0.066	0.027	NS
Old River, western arm at railroad bridge (902)	0.146	0.005	0.109	0.004	NS
San Joaquin River between Hog and Turner Cuts (910)	0.151	0.011	0.087	0.033	NS
Old River at mouth of Holland Cut (915)	0.162	0.013	0.077	0.012	Sig (48%)
Field Dup.: San Joaquin River between Hog and Turner Cuts (910)	0.143	0.011	-	-	NA

	MSD	PMSD
One-way ANOVA	0.070	82.2
Two-way ANOVA	0.074	86.3

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.
Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.
3. This low conductivity sample was compared to the low conductivity control.

Table A15-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 4/17/06.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Sacramento Deep Water Channel, Light 55	499	14.9	7.94	10.0	10.9	0.01	0.000
Sacramento R. across from Sherman Lake (704)	189	13.4	7.69	10.1	128.6	0.08	0.001
Sacramento River at tip of Grand Island (711)	100	11.8	7.75	11.0	32.4	0.07	0.001
Middle of Broad Slough, West end (804)	147	13.4	8.01	9.6	14.5	0.06	0.001
San Joaquin River, West of Oulton Point (812)	143	14.5	7.48	9.1	13.8	0.08	0.001
Old River, western arm at railroad bridge (902)	204	14.6	7.35	7.8	8.5	0.04	0.000
San Joaquin River between Hog and Turner Cuts (910)	176	14.6	7.27	7.6	9.9	0.13	0.001
Old River at mouth of Holland Cut (915)	190	14.6	7.24	7.4	9.6	0.10	0.000
Field Dup.: San Joaquin River between Hog and Turner Cuts (910)	176	14.6	7.27	7.6	9.9	0.12	0.001

Table A15-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 4/18/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 4/17/06.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	366	23.1	24.9	6.2	8.7	7.52	8.03	-	-	-
Low EC Control	124	23.4	25.0	6.5	8.6	7.30	7.81	-	-	-
Sacramento Deep Water Channel, Light 55	489	24.0	24.8	6.6	8.7	7.88	8.23	150	124	0.000
Sacramento R. across from Sherman Lake (704)	179	24.7	25.0	6.4	8.9	7.70	8.05	78	73	0.003
Sacramento River at tip of Grand Island (711)	116	24.9	25.4	6.7	9.4	7.42	7.84	52	47	0.001
Middle of Broad Slough, West end (804)	153	24.9	25.3	6.7	8.7	7.44	8.00	56	48	0.001
San Joaquin River, West of Oulton Point (812)	156	24.9	28.8	6.6	9.2	7.46	7.80	64	48	0.001
Old River, western arm at railroad bridge (902)	216	25.0	28.5	6.7	8.5	7.36	8.03	60	52	0.000
San Joaquin River between Hog and Turner Cuts (910)	191	25.0	29.6	6.8	8.8	7.31	7.86	64	50	0.001
Old River at mouth of Holland Cut (915)	199	25.0	28.5	6.8	8.6	7.36	7.92	60	48	0.001
Field Dup.: San Joaquin River between Hog and Turner Cuts (910)	207	24.4	24.7	6.8	8.6	7.45	7.94	60	50	0.002
DIEPAMHR + 100 ppb PBO	309	24.2	26.7	7.0	8.6	7.83	8.11	-	-	-
Low EC Control + 100 ppb PBO	132	23.9	25.5	6.9	8.8	7.49	7.98	-	-	-
Sacramento Deep Water Channel, Light 55 + 100 ppb PBO	492	23.9	26.4	6.9	8.7	7.99	8.14	-	-	-
Sacramento R. across from Sherman Lake (704) + 100 ppb PBO	184	24.2	27.5	6.3	8.7	7.66	8.08	-	-	-
Sacramento River at tip of Grand Island (711) + 100 ppb PBO	112	24.2	25.8	7.2	8.6	7.73	7.84	-	-	-
Middle of Broad Slough, West end (804) + 100 ppb PBO	164	24.3	25.7	7.1	8.7	7.55	8.00	-	-	-
San Joaquin River, West of Oulton Point (812) + 100 ppb PBO	157	23.9	25.2	7.2	9.0	7.57	7.81	-	-	-
Old River, western arm at railroad bridge (902) + 100 ppb PBO	212	23.9	24.1	7.2	8.7	7.47	8.02	-	-	-
San Joaquin River between Hog and Turner Cuts (910) + 100 ppb PBO	191	23.6	24.3	7.4	8.5	7.68	7.87	-	-	-
Old River at mouth of Holland Cut (915) + 100 ppb PBO	201.7	23.7	23.9	7.3	8.6	7.60	8.02	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A16-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 4/19/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 4/18/06.

Treatment	Survival (%) ¹				
	Unmanipulated		100 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	100	0.0	97	3.3	NS
San Pablo Bay at Rodeo Flats (323)	100	0.0	96	4.2	NS
Napa River at Vallejo Seawall (340)	100	0.0	100	0.0	NS
Carquinez Strait, West of Benicia army dock (405)	98	2.0	100	0.0	NS
Suisun Bay, East of middle point (504)	100	0.0	97	3.3	NS
Suisun Bay off Chipps Island (508)	82	18.0	86	3.2	NS
Grizzly Bay at Dolphin (602)	100	0.0	97	3.3	NS
Montezuma Slough at Nurse Slough (609)	100	0.0	100	0.0	NS
Trip Blank	84	16.0	97	3.3	NS

	MSD	PMSD
One-way ANOVA ³	-	-
Two-way ANOVA	39.0	39.0

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		100 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.125	0.009	0.084	0.010	NS
San Pablo Bay at Rodeo Flats (323)	0.168	0.015	0.122	0.023	NS
Napa River at Vallejo Seawall (340)	0.195	0.015	0.180	0.019	NS
Carquinez Strait, West of Benicia army dock (405)	0.139	0.015	0.159	0.012	NS
Suisun Bay, East of middle point (504)	0.162	0.012	0.162	0.012	NS
Suisun Bay off Chipps Island (508)	0.135	0.015	0.085	0.010	NS
Grizzly Bay at Dolphin (602)	0.164	0.024	0.152	0.005	NS
Montezuma Slough at Nurse Slough (609)	0.182	0.023	0.139	0.009	NS
Trip Blank	0.094	0.013	0.080	0.022	NS

	MSD	PMSD
One-way ANOVA	0.086	68.4
Two-way ANOVA	0.086	68.9

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. Survival in the unmanipulated samples was compared to the control using a Kruskal-Wallis test, and calculation of an MSD was not possible.

Table A16-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 4/18/06.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
San Pablo Bay at Rodeo Flats (323)	174	13.5	7.76	10.5	70.6	0.20	0.003
Napa River at Vallejo Seawall (340)	642	13.7	7.66	10.0	78.4	0.16	0.002
Carquinez Strait, West of Benicia army dock (405)	163	13.5	7.76	10.5	85.0	0.24	0.003
Suisun Bay, East of middle point (504)	156	12.9	7.76	10.6	83.8	0.17	0.002
Suisun Bay off Chipps Island (508)	157	12.9	7.76	10.4	83.4	0.16	0.002
Grizzly Bay at Dolphin (602)	159	13.1	7.74	10.6	83.5	0.16	0.002
Montezuma Slough at Nurse Slough (609)	305	12.8	7.91	10.2	109.2	0.20	0.003
Trip Blank	-	-	-	-	-	0.01	-

Table A16-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 4/19/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 4/18/06.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	361	20.0	24.7	7.2	8.1	7.67	8.13	88	60	-
San Pablo Bay at Rodeo Flats (323)	184	22.2	24.9	7.5	8.0	7.68	8.27	60	62	0.018
Napa River at Vallejo Seawall (340)	617	22.3	25.2	7.1	8.6	7.62	7.91	122	82	0.007
Carquinez Strait, West of Benicia army dock (405)	170	22.5	25.0	7.0	8.3	7.59	7.78	68	68	0.007
Suisun Bay, East of middle point (504)	171	22.5	25.0	7.1	8.1	7.61	7.97	60	66	0.008
Suisun Bay off Chipps Island (508)	172	22.7	25.1	7.0	8.1	7.57	7.97	64	66	0.008
Grizzly Bay at Dolphin (602)	186	23.0	25.0	6.8	8.0	7.68	8.07	72	80	0.009
Montezuma Slough at Nurse Slough (609)	288	23.3	25.1	7.1	8.5	7.69	7.95	92	74	0.009
Trip Blank	357	23.2	25.1	7.6	8.7	7.65	7.80	112	62	0.000
DIEPAMHR + 100 ppb PBO	373	23.2	24.6	7.5	8.1	7.75	8.15	-	-	-
San Pablo Bay at Rodeo Flats (323) + 100 ppb PBO	199	23.3	24.5	7.1	8.1	7.66	8.02	-	-	-
Napa River at Vallejo Seawall (340) + 100 ppb PBO	627	23.4	24.5	7.1	8.7	7.62	8.06	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 100 ppb PBO	185	23.6	24.5	7.1	8.1	7.61	8.07	-	-	-
Suisun Bay, East of middle point (504) + 100 ppb PBO	178	23.6	24.5	7.1	7.9	7.59	8.21	-	-	-
Suisun Bay off Chipps Island (508) + 100 ppb PBO	175	24.1	24.6	7.1	8.9	7.58	8.14	-	-	-
Grizzly Bay at Dolphin (602) + 100 ppb PBO	185	23.6	24.4	7.4	8.1	7.75	8.06	-	-	-
Montezuma Slough at Nurse Slough (609) + 100 ppb PBO	316	23.9	24.4	7.0	8.3	7.75	8.13	-	-	-
Trip Blank + 100 ppb PBO	373	24.3	24.4	7.4	8.7	7.75	8.11	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A17-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 5/03/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 5/01/06 - 5/02/06.

Treatment	Survival (%) ¹	
	Unmanipulated	
	mean	se
DIEPAMHR	98	2.0
Sacramento Deep Water Channel, Light 55	100	0.0
Napa River at Vallejo Seawall (340)	100	0.0
Carquinez Strait, West of Benicia army dock (405)	100	0.0
Suisun Bay off Chipps Island (508)	94	4.1
Sacramento River at tip of Grand Island (711)	94	4.2
Old River, western arm at railroad bridge (902)	100	0.0
San Joaquin River between Hog and Turner Cuts (910)	98	2.2
Old River at mouth of Holland Cut (915)	98	2.2
Field Dup.: Sacramento Deep Water Channel, Light 55	98	2.0

	MSD	PMSD
One-way ANOVA ³	-	-

Treatment	Weight (mg/surviving individual) ¹	
	Unmanipulated	
	mean	se
DIEPAMHR	0.071	0.004
Sacramento Deep Water Channel, Light 55	0.099	0.009
Napa River at Vallejo Seawall (340)	0.103	0.005
Carquinez Strait, West of Benicia army dock (405)	0.103	0.008
Suisun Bay off Chipps Island (508)	0.081	0.009
Sacramento River at tip of Grand Island (711)	0.079	0.002
Old River, western arm at railroad bridge (902)	0.113	0.004
San Joaquin River between Hog and Turner Cuts (910)	0.123	0.006
Old River at mouth of Holland Cut (915)	0.089	0.010
Field Dup.: Sacramento Deep Water Channel, Light 55	0.088	0.009

	MSD	PMSD
One-way ANOVA ³	-	-

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions showed extremely variable survival and weight, and were excluded from the analysis of this exposure.

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. Survival and weight in the unmanipulated samples were compared to the control using a Kruskal-Wallis test, and calculation of an MSD was not possible.

Table A17-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 5/01/06 - 5/02/06.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Sacramento Deep Water Channel, Light 55	500	18.5	8.2	10.2	12.2	0.03	0.001
Napa River at Vallejo Seawall (340)	846	17.2	7.83	8.4	53.7	0.07	0.001
Carquinez Strait, West of Benicia army dock (405)	530	17.4	7.93	9.2	66.7	0.06	0.001
Suisun Bay off Chipps Island (508)	135	16.9	7.46	7.6	5.4	0.04	0.000
Sacramento River at tip of Grand Island (711)	106	15.2	7.46	10.4	20.9	0.06	0.000
Old River, western arm at railroad bridge (902)	160	18.8	7.23	8.7	5.8	0.05	0.000
San Joaquin River between Hog and Turner Cuts (910)	141	18.3	7.21	8.6	7.8	0.08	0.000
Old River at mouth of Holland Cut (915)	86	18.2	7.19	8.1	7.2	0.04	0.000
Field Dup.: Sacramento Deep Water Channel, Light 55	500	18.5	8.2	10.2	12.2	0.02	0.001

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Table A17-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 5/03/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 5/01/06 - 5/02/06.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	321	22.3	24.7	7.5	8.4	7.86	8.23	110	59	-
Sacramento Deep Water Channel, Light 55	463	22.2	24.6	6.7	8.3	7.97	8.46	140	122	0.004
Napa River at Vallejo Seawall (340)	751	22.4	24.4	6.6	8.4	7.67	8.15	124	68	0.004
Carquinez Strait, West of Benicia army dock (405)	519	22.4	24.3	7.1	8.5	7.67	8.12	100	64	0.004
Suisun Bay off Chipps Island (508)	157	22.5	24.6	7.3	8.4	7.75	8.15	56	58	0.003
Sacramento River at tip of Grand Island (711)	116	22.6	24.8	7.1	8.4	7.71	8.16	52	46	0.004
Old River, western arm at railroad bridge (902)	138	22.6	24.1	6.5	8.5	7.60	7.80	56	40	0.001
San Joaquin River between Hog and Turner Cuts (910)	154	22.5	24.4	6.7	8.4	7.58	8.10	44	40	0.005
Old River at mouth of Holland Cut (915)	147	22.6	24.5	7.2	8.3	7.60	8.14	44	42	0.003
Field Dup.: Sacramento Deep Water Channel, Light 55	465	22.3	24.8	7.0	8.6	8.02	8.36	148	124	0.002

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A18-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 5/04/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 5/02/06 - 5/03/06.

Treatment	Survival (%) ¹				
	Unmanipulated		100 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	100	0.0	97	3.3	NS
San Pablo Bay at Rodeo Flats (323)	94	4.4	93	3.5	NS
Suisun Bay, East of middle point (504) ³	76	19.4	93	3.3	NS
Grizzly Bay at Dolphin (602) ³	71	16.3	97	3.0	NS
Montezuma Slough at Nurse Slough (609)	93	7.3	97	3.3	NS
Sacramento R. across from Sherman Lake (704)	85	5.7	83	8.8	NS
Middle of Broad Slough, West end (804) ³	94	2.4	33	28.5	NS
San Joaquin R., West of Oulton Point (812) ³	100	0.0	43	29.6	NS
Field Dup.: San Joaquin R., West of Oulton Point (812)	96	2.7	100	0.0	NS

	MSD	PMSD
One-way ANOVA	48.1	48.1
Two-way ANOVA	43.8	43.8

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		100 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.074	0.003	0.089	0.003	NS
San Pablo Bay at Rodeo Flats (323)	0.103	0.007	0.077	0.007	NS
Suisun Bay, East of middle point (504)	0.093	0.007	0.084	0.005	NS
Grizzly Bay at Dolphin (602)	0.109	0.010	0.089	0.004	NS
Montezuma Slough at Nurse Slough (609)	0.132	0.003	0.104	0.008	NS
Sacramento R. across from Sherman Lake (704)	0.096	0.004	0.054	0.006	NS
Middle of Broad Slough, West end (804)	0.094	0.007	0.052	0.012	NS
San Joaquin R., West of Oulton Point (812)	0.108	0.004	0.069	0.029	NS
Field Dup.: San Joaquin R., West of Oulton Point (812)	0.104	0.005	0.096	0.014	NS

	MSD	PMSD
One-way ANOVA	0.031	41.7
Two-way ANOVA	0.037	50.8

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. Some treatments showed lower mean survival due to very low survival in some replicates and high variability in survival between replicates. This pattern may be evidence of pathogen related mortality, and is not a clear indication of toxicity.

Table A18-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 5/02/06 - 5/03/06.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
San Pablo Bay at Rodeo Flats (323)	6016	16.5	7.69	8.8	38.0	0.08	0.001
Suisun Bay, East of middle point (504)	149	17.7	7.62	9.1	3.7	0.03	0.000
Grizzly Bay at Dolphin (602)	179	17.7	7.82	9.2	5.6	0.02	0.000
Montezuma Slough at Nurse Slough (609)	502	17.5	7.51	7.2	13.9	0.09	0.001
Sacramento R. across from Sherman Lake (704)	131	17.5	7.62	9.1	4.6	0.04	0.001
Middle of Broad Slough, West end (804)	129	18.0	7.82	8.6	-	0.06	0.001
San Joaquin R., West of Oulton Point (812)	116	17.8	7.58	7.1	4.1	0.08	0.001
Field Dup.: San Joaquin R., West of Oulton Point (812)	116	17.8	7.58	7.1	4.1	0.05	0.001

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Table A18-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 5/02/06 - 5/03/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 5/04/06.

Treatment	Laboratory Chemistry						Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹	
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH				Max pH
DIEPAMHR	415	23.4	24.7	6.1	8.3	7.74	8.08	110	59	-
San Pablo Bay at Rodeo Flats (323)	6555	23.3	24.8	7.1	8.3	7.76	7.78	772	70	0.002
Suisun Bay, East of middle point (504)	168	23.4	24.7	6.8	8.1	7.71	7.95	56	56	0.001
Grizzly Bay at Dolphin (602)	196	23.4	24.6	7.0	8.4	7.79	8.01	68	64	0.001
Montezuma Slough at Nurse Slough (609)	501	23.4	24.8	6.6	8.0	7.78	8.03	112	70	0.005
Sacramento R. across from Sherman Lake (704)	178	23.3	24.8	7.1	8.3	7.85	8.01	76	68	0.002
Middle of Broad Slough, West end (804)	152	23.3	24.8	6.8	8.3	7.61	7.82	48	46	0.002
San Joaquin R., West of Oulton Point (812)	142	23.2	24.6	6.4	8.1	7.58	7.89	48	44	0.003
Field Dup.: San Joaquin R., West of Oulton Point (812)	150	23.4	24.7	6.7	8.4	7.61	7.90	52	70	0.002
DIEPAMHR + 100 ppb PBO	334	23.4	24.4	6.7	8.1	7.83	8.07	-	-	-
San Pablo Bay at Rodeo Flats (323) + 100 ppb PBO	6580	23.3	24.4	7.1	8.2	7.67	7.77	-	-	-
Field Dup.: San Joaquin R., West of Oulton Point (812) + 100 ppb PBO	142	23.2	24.4	6.7	8.6	7.61	7.87	-	-	-
Suisun Bay, East of middle point (504) + 100 ppb PBO	175	23.3	24.4	7.1	8.4	7.79	7.96	-	-	-
Grizzly Bay at Dolphin (602) + 100 ppb PBO	201	23.2	24.4	6.9	8.2	7.78	8.05	-	-	-
Montezuma Slough at Nurse Slough (609) + 100 ppb PBO	550	23.2	24.4	7.1	8.2	7.89	7.97	-	-	-
Sacramento R. across from Sherman Lake (704) + 100 ppb PBO	184	23.2	24.3	7.1	8.6	7.84	8.06	-	-	-
Middle of Broad Slough, West end (804) + 100 ppb PBO	147	23.2	24.4	6.9	8.3	7.67	7.81	-	-	-
San Joaquin R., West of Oulton Point (812) + 100 ppb PBO	147	23.3	24.3	6.8	8.2	7.71	7.76	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A19-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 5/17/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 5/15/06 - 5/16/06.

Treatment	Survival (%) ¹				
	Unmanipulated		50 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	96	2.4	73	16.7	NS
High EC DIEPAMHR @ 11.53 mS/cm	98	2.0	89	5.9	NS
Sacramento River at tip of Grand Island (711)	82	6.3	60	14.9	NS
San Joaquin River between Hog and Turner Cuts (910)	96	2.4	67	33.3	NS
Old River at mouth of Holland Cut (915)	98	2.2	80	20.0	NS
Napa River at Riverside Blvd. terminus (340)	100	0.0	91	9.1	NS
Suisun Bay off Chipps Island (508)	85	9.0	90	10.0	NS
Montezuma Slough at Nurse Slough (609)	98	2.0	100	0.0	NS
Sacramento Deep Water Channel, Light 55	92	3.7	93	6.7	NS
San Pablo Bay at Rodeo Flats (323)	98	2.2	100	0.0	NS
Carquinez Strait, West of Benicia army dock (405)	98	2.0	97	3.3	NS
Old River, western arm at railroad bridge (902)	66	19.1	97	3.3	NS
Field Dup.: Sacramento Deep Water Channel, Light 55	100	0.0	-	-	NA

	MSD	PMSD
One-way ANOVA ³	-	-
Two-way ANOVA	51.9	54.0

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		50 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.055	0.010	0.030	0.012	NS
High EC DIEPAMHR @ 11.53 mS/cm	0.073	0.012	0.020	0.006	NS
Sacramento River at tip of Grand Island (711)	0.058	0.007	0.106	0.012	NS
San Joaquin River between Hog and Turner Cuts (910)	0.099	0.008	0.106	0.008	NS
Old River at mouth of Holland Cut (915)	0.116	0.014	0.115	0.014	NS
Napa River at Riverside Blvd. terminus (340)	0.102	0.005	0.111	0.026	NS
Suisun Bay off Chipps Island (508)	0.078	0.010	0.107	0.010	NS
Montezuma Slough at Nurse Slough (609)	0.102	0.008	0.114	0.016	NS
Sacramento Deep Water Channel, Light 55	0.088	0.009	0.130	0.006	NS
San Pablo Bay at Rodeo Flats (323)	0.067	0.011	0.065	0.015	NS
Carquinez Strait, West of Benicia army dock (405)	0.072	0.011	0.133	0.005	NS
Old River, western arm at railroad bridge (902)	0.068	0.011	0.088	0.006	NS
Field Dup.: Sacramento Deep Water Channel, Light 55	0.095	0.008	-	-	NA

	MSD	PMSD
One-way ANOVA	0.054	102.8
Two-way ANOVA	0.063	120.1

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. Survival and weight in the unmanipulated samples were compared to the control using a Kruskal-Wallis test, and calculation of an MSD was not possible.

Table A19-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 5/15/06 - 5/16/06.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Sacramento River at tip of Grand Island (711)	96	17.2	7.21	9.4	29.0	0.09	0.000
San Joaquin River between Hog and Turner Cuts (910)	128	20.6	7.16	7.2	8.5	0.14	0.001
Old River at mouth of Holland Cut (915)	126	20.1	7.10	7.8	10.9	0.10	0.000
Napa River at Riverside Blvd. terminus (340)	2368	18.9	7.78	7.9	63.1	0.10	0.002
Suisun Bay off Chipps Island (508)	121	20.7	7.58	7.6	13.4	0.11	0.002
Montezuma Slough at Nurse Slough (609)	318	19.8	7.56	7.8	74.0	0.13	0.002
Sacramento Deep Water Channel, Light 55	534	22.7	8.07	7.7	22.4	0.11	0.005
San Pablo Bay at Rodeo Flats (323)	11500	18.0	7.84	7.7	95.8	0.19	0.003
Carquinez Strait, West of Benicia army dock (405)	230	19.1	7.94	7.6	80.1	0.08	0.002
Old River, western arm at railroad bridge (902)	190	20.6	6.99	7.1	9.3	0.04	0.000
Field Dup.: Sacramento Deep Water Channel, Light 55	534	22.7	8.07	7.7	22.4	0.11	0.005

Table A19-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 5/17/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 5/15/06 - 5/16/06.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	325	23.1	24.3	6.5	8.5	7.58	7.82	110	57	-
High EC DIEPAMHR @ 11.53 mS/cm	5773	22.9	24.2	7.6	8.9	7.44	7.78	736	212	-
Sacramento River at tip of Grand Island (711)	186	23.1	24.3	7.2	8.9	7.61	7.81	44	46	0.003
San Joaquin River between Hog and Turner Cuts (910)	152	23.2	24.4	7.0	8.7	7.00	7.72	44	37	0.004
Old River at mouth of Holland Cut (915)	150	23.3	24.5	6.9	8.8	7.45	7.62	36	35	0.002
Napa River at Riverside Blvd. terminus (340)	14123	23.3	24.5	7.0	8.7	7.53	7.62	280	72	0.002
Suisun Bay off Chipps Island (508)	161	23.4	24.4	6.5	8.5	7.48	7.68	52	46	0.003
Montezuma Slough at Nurse Slough (609)	343	23.6	24.6	6.6	8.8	7.59	7.70	72	58	0.003
Sacramento Deep Water Channel, Light 55	543	23.5	24.3	6.8	8.9	7.28	8.04	164	140	0.004
San Pablo Bay at Rodeo Flats (323)	11340	23.7	24.2	6.8	8.6	7.32	7.63	1376	68	0.002
Carquinez Strait, West of Benicia army dock (405)	240	23.8	24.1	6.8	8.9	7.80	7.95	1032	54	0.003
Old River, western arm at railroad bridge (902)	158	23.7	23.8	6.8	8.7	7.66	7.71	44	38	0.001
Field Dup.: Sacramento Deep Water Channel, Light 55	559	23.7	24.5	6.8	8.7	7.81	8.05	168	138	0.004
DIEPAMHR + 50 ppb PBO	2018	24.3	25.8	6.6	8.4	7.51	8.14	-	-	-
High EC DIEPAMHR @ 11.53 mS/cm + 50 ppb PBO	11680	24.3	25.7	7.2	8.9	7.19	8.03	-	-	-
Sacramento River at tip of Grand Island (711) + 50 ppb PBO	135	24.6	25.7	6.8	8.4	7.28	8.05	-	-	-
San Joaquin River between Hog and Turner Cuts (910) + 50 ppb PBO	158	24.3	25.6	6.9	8.5	7.60	7.96	-	-	-
Old River at mouth of Holland Cut (915) + 50 ppb PBO	157	24.3	25.7	7.0	8.8	7.47	7.99	-	-	-
Napa River at Riverside Blvd. terminus (340) + 50 ppb PBO	2452	24.4	25.5	7.1	8.7	7.62	7.84	-	-	-
Suisun Bay off Chipps Island (508) + 50 ppb PBO	150	24.4	25.5	7.0	8.5	7.51	8.05	-	-	-
Montezuma Slough at Nurse Slough (609) + 50 ppb PBO	353.85	24.3	25.4	6.9	8.9	7.80	8.07	-	-	-
Sacramento Deep Water Channel, Light 55 + 50 ppb PBO	583	24.7	25.4	6.7	8.5	7.62	8.21	-	-	-
San Pablo Bay at Rodeo Flats (323) + 50 ppb PBO	11810	24.6	25.4	6.8	8.8	7.61	7.85	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 50 ppb PBO	220	24.4	25.3	6.9	8.8	7.55	8.25	-	-	-
Old River, western arm at railroad bridge (902) + 50 ppb PBO	157	24.7	25.3	6.7	8.7	7.47	8.11	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A20-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 5/18/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 5/17/06.

Treatment	Survival (%) ¹				
	Unmanipulated		50 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	96	2.4	93	3.3	NS
Suisun Bay, East of middle point (504)	92	3.7	100	0.0	NS
Grizzly Bay at Dolphin (602)	96	4.0	97	3.3	NS
Sacramento R. across from Sherman Lake (704)	90	5.5	82	7.8	NS
Middle of Broad Slough, West end (804)	98	2.0	100	0.0	NS
San Joaquin River, West of Oulton Point (812)	98	1.8	91	5.3	NS
Bottle Blank	98	2.0	-	-	NA

	MSD	PMSD
One-way ANOVA	16.7	17.4
Two-way ANOVA	19.0	19.8

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		50 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.054	0.002	0.049	0.006	NS
Suisun Bay, East of middle point (504)	0.099	0.013	0.079	0.007	NS
Grizzly Bay at Dolphin (602)	0.113	0.013	0.119	0.011	NS
Sacramento R. across from Sherman Lake (704)	0.086	0.014	0.069	0.006	NS
Middle of Broad Slough, West end (804)	0.098	0.006	0.076	0.008	NS
San Joaquin River, West of Oulton Point (812)	0.111	0.009	0.067	0.006	NS
Bottle Blank	0.070	0.003	-	-	NA

	MSD	PMSD
One-way ANOVA	0.049	91.2
Two-way ANOVA	0.050	93.5

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

Table A20-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 5/17/06.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Suisun Bay, East of middle point (504)	123	17.8	7.81	8.8	8.3	0.03	0.001
Grizzly Bay at Dolphin (602)	153	18.5	7.5	8.3	27.2	0.13	0.001
Sacramento R. across from Sherman Lake (704)	107	17.4	7.81	9.6	15.6	0.06	0.001
Middle of Broad Slough, West end (804)	114	20.0	7.61	8.2	29.0	0.05	0.001
San Joaquin River, West of Oulton Point (812)	107	19.5	7.61	8.3	6.4	0.06	0.001
Bottle Blank	294	22.9	7.92	8.3	0.4	0.00	0.000

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Table A20-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 5/18/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 5/17/06.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	326	23.2	24.8	6.5	8.3	7.46	8.12	110	57	-
Suisun Bay, East of middle point (504)	148	24.7	26.8	6.4	8.4	7.43	7.93	88	48	0.001
Grizzly Bay at Dolphin (602)	180	23.5	24.7	6.8	8.5	7.49	8.00	52	50	0.006
Sacramento R. across from Sherman Lake (704)	132	23.1	24.5	6.8	8.9	7.45	7.93	46	51	0.002
Middle of Broad Slough, West end (804)	136	23.3	24.4	6.2	8.2	7.36	7.95	38	37	0.002
San Joaquin River, West of Oulton Point (812)	131	23.2	24.4	6.4	8.4	7.44	7.94	42	38	0.002
Bottle Blank	328	23.4	24.7	6.8	8.6	7.45	7.97	98	56	0.000
DIEPAMHR + 50 ppb PBO	324	23.3	24.1	6.5	8.2	7.43	8.13	-	-	-
Suisun Bay, East of middle point (504) + 50 ppb PBO	146	23.3	24.0	6.4	8.5	7.50	7.99	-	-	-
Grizzly Bay at Dolphin (602) + 50 ppb PBO	179	23.3	24.0	6.7	8.4	7.52	8.02	-	-	-
Sacramento R. across from Sherman Lake (704) + 50 ppb PBO	137	23.3	23.9	6.9	8.4	7.54	7.83	-	-	-
Middle of Broad Slough, West end (804) + 50 ppb PBO	137	23.3	24.0	6.2	8.3	7.44	7.87	-	-	-
San Joaquin River, West of Oulton Point (812) + 50 ppb PBO	136	23.2	23.9	6.7	8.3	7.54	7.86	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A21-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 6/1/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 5/30/06 - 5/31/06.

Treatment	Survival (%) ¹				
	Unmanipulated		50 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	94	2.5	93	6.7	NS
High EC Control @ 12.30 mS/cm	90	3.2	-	-	NA
Sacramento Deep Water Channel, Light 55	98	2.0	91	5.8	NS
Sacramento River at tip of Grand Island (711)	90	3.2	87	3.6	NS
Old River, western arm at railroad bridge (902)	91	4.2	100	0.0	NS
San Joaquin River between Hog and Turner Cuts (910)	89	4.0	94	3.2	NS
Old River at mouth of Holland Cut (915)	90	5.5	83	6.7	NS
San Pablo Bay at Rodeo Flats (323)	96	3.6	100	0.0	NS
Napa River at Vallejo Seawall (340)	94	4.0	100	0.0	NS
Carquinez Strait, West of Benicia army dock (405)	100	0.0	97	3.0	NS
Suisun Bay off Chipps Island (508)	80	11.9	80	0.0	NS
Montezuma Slough at Nurse Slough (609)	98	2.2	97	3.3	NS

	MSD	PMSD
One-way ANOVA ³	-	-
Two-way ANOVA	8.2	8.7

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		50 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.066	0.008	0.067	0.006	NS
High EC Control @ 12.30 mS/cm	0.047	0.011	-	-	NA
Sacramento Deep Water Channel, Light 55	0.114	0.011	0.082	0.008	NS
Sacramento River at tip of Grand Island (711)	0.046	0.008	0.058	0.010	NS
Old River, western arm at railroad bridge (902)	0.072	0.006	0.065	0.002	NS
San Joaquin River between Hog and Turner Cuts (910)	0.071	0.005	0.064	0.010	NS
Old River at mouth of Holland Cut (915)	0.082	0.015	0.058	0.013	NS
San Pablo Bay at Rodeo Flats (323)	0.069	0.007	0.052	0.007	NS
Napa River at Vallejo Seawall (340)	0.083	0.006	0.082	0.010	NS
Carquinez Strait, West of Benicia army dock (405)	0.096	0.013	0.075	0.010	NS
Suisun Bay off Chipps Island (508)	0.086	0.017	0.089	0.037	NS
Montezuma Slough at Nurse Slough (609)	0.097	0.008	0.093	0.028	NS

	MSD	PMSD
One-way ANOVA	0.056	84.5
Two-way ANOVA	0.066	100.3

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. Survival and weight in the unmanipulated samples were compared to the control using a Kruskal-Wallis test, and calculation of an MSD was not possible.

Table A21-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 5/30/06 - 5/31/06.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Sacramento Deep Water Channel, Light 55	389	19.4	7.89	7.7	26.5	0.11	0.003
Sacramento River at tip of Grand Island (711)	109	17.2	7.53	7.7	21.1	0.14	0.001
Old River, western arm at railroad bridge (902)	186	19.1	6.98	8.1	8.9	0.09	0.000
San Joaquin River between Hog and Turner Cuts (910)	115	18.3	7.37	8.4	10.5	0.14	0.001
Old River at mouth of Holland Cut (915)	123	18.3	7.19	8.9	9.3	0.10	0.001
San Pablo Bay at Rodeo Flats (323)	11660	17.3	7.41	8.4	122.0	0.12	0.001
Napa River at Vallejo Seawall (340)	2875	18.1	7.61	8.6	49.0	0.09	0.001
Carquinez Strait, West of Benicia army dock (405)	249	17.8	7.51	8.1	79.1	0.09	0.001
Suisun Bay off Chipps Island (508)	100	18.2	7.51	7.8	14.2	0.06	0.001
Montezuma Slough at Nurse Slough (609)	591	19.2	7.28	8.5	56.1	0.11	0.001

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Table A21-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 6/01/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 5/30/06 - 5/31/06.

Treatment	Laboratory Chemistry						Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹	
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH				Max pH
DIEPAMHR	371	22.1	24.0	6.6	8.1	7.69	8.19	102	62	-
High EC Control @ 12.30 mS/cm	11910	23.5	23.8	6.6	8.3	7.56	8.03	-	-	-
Sacramento Deep Water Channel, Light 55	427	24.0	24.1	6.0	8.3	7.85	8.09	126	112	0.006
Sacramento River at tip of Grand Island (711)	141	23.9	24.8	6.1	8.3	7.51	8.05	50	52	0.007
Old River, western arm at railroad bridge (902)	164	23.7	24.1	6.0	8.5	7.44	7.85	40	35	0.003
San Joaquin River between Hog and Turner Cuts (910)	150	23.8	24.2	6.4	8.4	7.34	7.86	40	35	0.005
Old River at mouth of Holland Cut (915)	165	23.7	24.2	6.1	8.6	7.45	7.68	32	34	0.002
San Pablo Bay at Rodeo Flats (323)	11895	23.6	24.1	6.5	8.1	7.36	7.67	1298	62	0.002
Napa River at Vallejo Seawall (340)	3375	23.8	24.3	6.3	8.3	7.54	7.84	382	61	0.003
Carquinez Strait, West of Benicia army dock (405)	283	24.0	24.2	5.9	8.4	7.55	7.91	58	49	0.004
Suisun Bay off Chipps Island (508)	161	23.6	24.1	5.9	8.9	7.58	7.62	45	46	0.001
Montezuma Slough at Nurse Slough (609)	614	23.7	24.2	6.4	8.1	7.55	7.92	92	60	0.004
DIEPAMHR + 50 ppb PBO	366	23.6	24.0	6.2	8.1	7.75	8.19	-	-	-
Sacramento Deep Water Channel, Light 55 + 50 ppb PBO	412	23.2	24.0	6.3	8.3	7.85	8.09	-	-	-
Sacramento River at tip of Grand Island (711) + 50 ppb PBO	137	23.8	23.9	6.0	8.3	7.66	8.05	-	-	-
Old River, western arm at railroad bridge (902) + 50 ppb PBO	162	23.2	24.1	6.3	8.5	7.53	7.85	-	-	-
San Joaquin River between Hog and Turner Cuts (910) + 50 ppb PBO	144	23.3	24.2	6.7	8.4	7.63	7.86	-	-	-
Old River at mouth of Holland Cut (915) + 50 ppb PBO	160	22.9	24.2	6.4	8.6	7.49	7.69	-	-	-
San Pablo Bay at Rodeo Flats (323) + 50 ppb PBO	11780	23.2	24.1	6.9	8.1	7.36	7.67	-	-	-
Napa River at Vallejo Seawall (340) + 50 ppb PBO	3305	23.0	24.3	6.0	8.3	7.55	7.89	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 50 ppb PBO	278.6	24.0	24.2	5.9	8.4	7.67	7.91	-	-	-
Suisun Bay off Chipps Island (508) + 50 ppb PBO	156	23.5	24.2	5.7	8.9	7.50	7.61	-	-	-
Montezuma Slough at Nurse Slough (609) + 50 ppb PBO	609	23.6	23.8	6.1	8.1	7.64	7.92	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A22-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 6/02/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 6/01/06.

Treatment	Survival (%) ¹				
	Unmanipulated		50 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	100	0.0	96	3.7	NS
Suisun Bay, East of middle point (504)	89	6.1	83	8.8	NS
Grizzly Bay at Dolphin (602)	98	2.0	100	0.0	NS
Sacramento R. across from Sherman Lake (704)	94	4.0	87	6.4	NS
Middle of Broad Slough, West end (804)	92	3.7	83	11.9	NS
San Joaquin River, West of Oulton Point (812)	90	6.3	90	0.0	NS
Field Dup.: Middle of Broad Slough, West end (804)	80	6.1	83	8.8	NS
Bottle Blank	94	4.4	-	-	NA

	MSD	PMSD
One-way ANOVA ³	-	-
Two-way ANOVA	27.3	27.3

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		50 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.057	0.003	0.053	0.008	NS
Suisun Bay, East of middle point (504)	0.055	0.007	0.073	0.012	NS
Grizzly Bay at Dolphin (602)	0.074	0.007	0.067	0.003	NS
Sacramento R. across from Sherman Lake (704)	0.079	0.010	0.070	0.005	NS
Middle of Broad Slough, West end (804)	0.076	0.009	0.065	0.014	NS
San Joaquin River, West of Oulton Point (812)	0.053	0.005	0.063	0.013	NS
Field Dup.: Middle of Broad Slough, West end (804)	0.059	0.009	0.061	0.005	NS
Bottle Blank	0.051	0.010	-	-	NA

	MSD	PMSD
One-way ANOVA	0.041	70.9
Two-way ANOVA	0.043	74.4

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. Survival and weight in the unmanipulated samples were compared to the control using a Kruskal-Wallis test, and calculation of an MSD was not possible.

Table A22-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 6/01/06.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Suisun Bay, East of middle point (504)	130	18.7	7.4	8.5	19.5	0.04	0.000
Grizzly Bay at Dolphin (602)	183	19.0	7.71	9.6	37.4	0.05	0.001
Sacramento R. across from Sherman Lake (704)	131	18.1	7.28	8.2	12.1	0.07	0.000
Middle of Broad Slough, West end (804)	132	18.1	7.68	8.2	7.3	0.03	0.000
San Joaquin River, West of Oulton Point (812)	110	18.9	7.28	8.6	9.2	0.08	0.001
Field Dup.: Middle of Broad Slough, West end (804)	132	18.1	7.68	8.2	7.1	0.04	0.001
Bottle Blank	355	23	8.14	9.1	0.1	0.00	0.000

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Table A22-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 6/02/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 6/01/06.

Treatment	Laboratory Chemistry						Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹	
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH				Max pH
DIEPAMHR	375	22.5	24.1	5.6	8.2	7.44	8.14	102	62	-
Suisun Bay, East of middle point (504)	161	22.4	23.7	5.7	8.3	7.34	8.08	46	50	0.002
Grizzly Bay at Dolphin (602)	213	22.2	23.8	5.5	8.4	7.32	7.94	56	52	0.002
Sacramento R. across from Sherman Lake (704)	170	22.6	23.7	5.7	8.2	7.35	8.08	54	54	0.004
Middle of Broad Slough, West end (804)	145	22.4	23.6	5.5	8.3	7.24	7.93	38	40	0.001
San Joaquin River, West of Oulton Point (812)	138	22.7	23.5	5.8	8.2	7.28	7.81	36	36	0.002
Field Dup.: Middle of Broad Slough, West end (804)	165	22.2	23.9	5.7	8.4	7.25	8.00	38	35	0.002
Bottle Blank	371	22.3	23.8	5.4	8.3	7.35	8.17	106	62	0.000
DIEPAMHR + 50 ppb PBO	370	22.7	24.0	5.9	8.3	7.43	8.17	-	-	-
Suisun Bay, East of middle point (504) + 50 ppb PBO	160	22.7	23.4	5.9	8.9	7.33	7.95	-	-	-
Grizzly Bay at Dolphin (602) + 50 ppb PBO	207	23.0	23.3	5.5	8.9	7.32	7.93	-	-	-
Sacramento R. across from Sherman Lake (704) + 50 ppb PBO	172	23.2	23.6	5.2	8.7	7.75	7.99	-	-	-
Middle of Broad Slough, West end (804) + 50 ppb PBO	148	23.2	23.6	5.8	8.6	7.30	7.86	-	-	-
San Joaquin River, West of Oulton Point (812) + 50 ppb PBO	140	23.2	23.2	6.2	8.9	7.30	7.99	-	-	-
Field Dup.: Middle of Broad Slough, West end (804) + 50 ppb PBO	150	22.8	23.6	5.9	8.4	7.24	7.89	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A23-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 6/15/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 6/13/06 - 6/14/06.

Treatment	Survival (%) ¹				
	Unmanipulated		50 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	98	2.0	100	0.0	NS
High EC Control @ 20.00 mS/cm	100	0.0	-	-	NA
San Joaquin River between Hog and Turner Cuts (910)	100	0.0	100	0.0	NS
Old River at mouth of Holland Cut (915)	100	0.0	100	0.0	NS
Old River, western arm at railroad bridge (902)	98	2.0	70	30.0	NS
Sacramento River at tip of Grand Island (711)	88	4.0	87	3.3	NS
Sacramento Deep Water Channel, Light 55	96	4.0	100	0.0	NS
San Pablo Bay at Rodeo Flats (323)	94	2.5	97	3.3	NS
Carquinez Strait, West of Benicia army dock (405)	100	0.0	100	0.0	NS
Napa River at Vallejo Seawall (340)	100	0.0	100	0.0	NS
Suisun Bay off Chipps Island (508)	92	3.5	93	3.3	NS
Montezuma Slough at Nurse Slough (609)	98	2.0	97	3.3	NS
San Joaquin River, West of Oulton Point (812)	100	0.0	94	3.2	NS

	MSD	PMSD
One-way ANOVA ³	-	-
Two-way ANOVA	26.6	27.2

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		50 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.075	0.007	0.084	0.006	NS
High EC Control @ 20.00 mS/cm	0.083	0.003	-	-	NA
San Joaquin River between Hog and Turner Cuts (910)	0.102	0.007	0.124	0.005	NS
Old River at mouth of Holland Cut (915)	0.116	0.004	0.129	0.015	NS
Old River, western arm at railroad bridge (902)	0.104	0.004	0.098	0.008	NS
Sacramento River at tip of Grand Island (711)	0.077	0.010	0.077	0.007	NS
Sacramento Deep Water Channel, Light 55	0.099	0.006	0.158	0.007	NS
San Pablo Bay at Rodeo Flats (323)	0.047	0.007	0.063	0.016	NS
Carquinez Strait, West of Benicia army dock (405)	0.102	0.006	0.127	0.018	NS
Napa River at Vallejo Seawall (340)	0.099	0.008	0.119	0.013	NS
Suisun Bay off Chipps Island (508)	0.080	0.009	0.085	0.018	NS
Montezuma Slough at Nurse Slough (609)	0.094	0.011	0.068	0.030	NS
San Joaquin River, West of Oulton Point (812)	0.069	0.008	0.120	0.012	NS

	MSD	PMSD
One-way ANOVA	0.040	53.7
Two-way ANOVA	0.050	66.6

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. Survival in the unmanipulated samples was compared to the control using a Kruskal-Wallis test, and calculation of an MSD was not possible.

Table A23-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 6/13/06 - 6/14/06.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
San Joaquin River between Hog and Turner Cuts (910)	121	20.4	7.61	9.0	7.8	0.10	0.002
Old River at mouth of Holland Cut (915)	173	22.1	7.28	8.1	5.9	0.06	0.000
Old River, western arm at railroad bridge (902)	132	21.7	7.12	10.2	3.3	0.07	0.000
Sacramento River at tip of Grand Island (711)	114	19.0	7.62	7.0	3.0	0.37	0.005
Sacramento Deep Water Channel, Light 55	425	20.7	7.6	7.8	13.8	0.02	0.000
San Pablo Bay at Rodeo Flats (323)	15650	17.9	7.4	8.3	219.7	0.16	0.001
Carquinez Strait, West of Benicia army dock (405)	1544	18.6	7.19	9.0	24.5	0.18	0.001
Napa River at Vallejo Seawall (340)	7580	17.7	7.41	8.2	30.4	0.14	0.001
Suisun Bay off Chipps Island (508)	133	19.4	7.41	8.1	9.2	0.13	0.001
Montezuma Slough at Nurse Slough (609)	188	19.7	7.61	7.2	13.8	0.06	0.001
San Joaquin River, West of Oulton Point (812)	120	21.0	7.81	7.9	4.4	0.08	0.002

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Table A23-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 6/15/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 6/13/06 - 6/14/06.

Treatment	Laboratory Chemistry						Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹	
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH				Max pH
DIEPAMHR	379	24.6	25.2	6.0	8.0	7.78	8.13	108	60	-
High EC Control @ 20.00 mS/cm	10178	24.1	26.1	6.5	8.9	7.54	8.22	-	-	-
San Joaquin River between Hog and Turner Cuts (910)	147	24.8	25.0	5.6	8.7	7.67	8.33	42	32	0.006
Old River at mouth of Holland Cut (915)	196	24.8	25.1	5.8	8.5	7.55	8.25	52	36	0.003
Old River, western arm at railroad bridge (902)	159	24.9	25.0	5.5	8.4	7.52	8.30	40	38	0.004
Sacramento River at tip of Grand Island (711)	139	24.8	24.8	5.6	8.4	7.54	8.33	48	48	0.021
Sacramento Deep Water Channel, Light 55	453	24.7	24.7	6.2	8.2	7.85	8.28	130	113	0.001
San Pablo Bay at Rodeo Flats (323)	18065	24.5	24.6	6.2	8.4	7.49	7.89	2320	77	0.002
Carquinez Strait, West of Benicia army dock (405)	1578	24.6	24.7	6.1	8.7	7.63	8.26	188	54	0.009
Napa River at Vallejo Seawall (340)	7410	24.7	24.7	6.2	8.5	7.56	8.05	876	70	0.004
Suisun Bay off Chipps Island (508)	166	24.7	24.8	5.9	8.1	7.64	8.42	44	48	0.009
Montezuma Slough at Nurse Slough (609)	215	24.5	24.6	6.0	8.8	7.62	8.34	60	52	0.004
San Joaquin River, West of Oulton Point (812)	157	24.4	26.4	5.9	8.8	7.54	8.32	46	39	0.005
DIEPAMHR + 50 ppb PBO	372	24.3	27.1	6.1	8.1	7.76	8.21	-	-	-
San Joaquin River between Hog and Turner Cuts (910) + 50 ppb PBO	150	24.2	25.1	6.2	8.6	7.48	8.24	-	-	-
Old River at mouth of Holland Cut (915) + 50 ppb PBO	200	24.1	25.7	6.0	8.3	7.53	8.22	-	-	-
Old River, western arm at railroad bridge (902) + 50 ppb PBO	159	23.8	25.0	5.8	8.6	7.55	8.30	-	-	-
Sacramento River at tip of Grand Island (711) + 50 ppb PBO	144	23.6	25.4	6.2	8.3	7.56	8.30	-	-	-
Sacramento Deep Water Channel, Light 55 + 50 ppb PBO	455	23.5	25.8	6.0	8.4	7.96	8.26	-	-	-
San Pablo Bay at Rodeo Flats (323) + 50 ppb PBO	18545	23.5	25.6	5.9	8.4	7.56	7.85	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 50 ppb PBO	1600	24.0	25.6	6.4	8.3	7.59	8.16	-	-	-
Napa River at Vallejo Seawall (340) + 50 ppb PBO	7485	23.7	25.8	5.9	8.2	7.53	8.05	-	-	-
Suisun Bay off Chipps Island (508) + 50 ppb PBO	176	23.9	25.6	6.2	8.3	7.58	8.23	-	-	-
Montezuma Slough at Nurse Slough (609) + 50 ppb PBO	219	23.6	25.4	5.9	8.2	7.57	8.11	-	-	-
San Joaquin River, West of Oulton Point (812) + 50 ppb PBO	156	23.3	25.9	5.8	8.4	7.52	8.40	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A24-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 6/16/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 6/15/06.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	96	2.4	100	0.0	NS
Suisun Bay, East of middle point (504)	100	0.0	97	3.3	NS
Grizzly Bay at Dolphin (602)	100	0.0	100	0.0	NS
Sacramento R. across from Sherman Lake (704)	90	4.3	100	0.0	NS
Middle of Broad Slough, West end (804)	94	2.4	100	0.0	NS
Field Dup.:Middle of Broad Slough, West end (804)	98	2.0	100	0.0	NS

	MSD	PMSD
One-way ANOVA ³	-	-
Two-way ANOVA	11.3	11.8

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.097	0.005	0.082	0.002	NS
Suisun Bay, East of middle point (504)	0.148	0.008	0.112	0.001	NS
Grizzly Bay at Dolphin (602)	0.135	0.012	0.118	0.010	NS
Sacramento R. across from Sherman Lake (704)	0.125	0.009	0.114	0.007	NS
Middle of Broad Slough, West end (804)	0.112	0.006	0.113	0.003	NS
Field Dup.:Middle of Broad Slough, West end (804)	0.109	0.008	0.140	0.012	NS

	MSD	PMSD
One-way ANOVA ³	-	-
Two-way ANOVA	0.041	42.0

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. Survival and weight in the unmanipulated samples were compared to the control using Kruskal-Wallis tests, and calculations of MSDs were not possible.

Table A24-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 6/15/06.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Suisun Bay, East of middle point (504)	198	19.1	7.81	7.9	9.9	0.08	0.002
Grizzly Bay at Dolphin (602)	145	19.1	7.64	8.6	13.7	0.04	0.001
Sacramento R. across from Sherman Lake (704)	127	19.1	7.28	7.9	6.7	0.07	0.000
Middle of Broad Slough, West end (804)	119	20.2	7.51	7.7	6.3	0.08	0.001
Field Dup.:Middle of Broad Slough, West end (804)	119	20.2	7.51	7.7	6.0	0.08	0.001

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Table A24-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 6/16/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 6/15/06.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPMAHR	364	24.5	25.1	5.4	8.2	7.61	8.06	108	60	-
Suisun Bay, East of middle point (504)	220	24.7	25.1	5.2	8.2	7.39	8.01	54	50	0.004
Grizzly Bay at Dolphin (602)	176	24.6	25.1	5.5	8.7	7.51	7.82	56	48	0.001
Sacramento R. across from Sherman Lake (704)	159	24.6	25.1	5.4	8.2	7.42	7.97	48	48	0.003
Middle of Broad Slough, West end (804)	153	25.0	25.0	5.4	8.4	7.51	7.91	40	40	0.003
Field Dup.:Middle of Broad Slough, West end (804)	150	24.8	25.1	5.3	8.6	7.59	7.80	44	41	0.003
DIEPAMHR + 25 ppb PBO	362	24.7	25.4	6.0	8.1	7.67	8.06	-	-	-
Suisun Bay, East of middle point (504) + 25 ppb PBO	221	24.7	25.6	5.6	8.4	7.62	7.91	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	176	24.6	25.3	5.1	8.4	7.48	7.98	-	-	-
Sacramento R. across from Sherman Lake (704) + 25 ppb PBO	158	24.6	25.1	5.2	8.4	7.56	7.89	-	-	-
Middle of Broad Slough, West end (804) + 25 ppb PBO	150	24.6	25.3	5.2	8.5	7.44	7.89	-	-	-
Field Dup.:Middle of Broad Slough, West end (804) + 25 ppb PBO	156	24.6	25.3	5.1	8.3	7.49	7.93	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A25-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 6/29/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 6/27/06 - 6/28/06.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	98	2.0	100	0.0	NS
High EC Control @ 21.9 mS/cm	89	3.5	62	4.3	NS
Sacramento River at tip of Grand Island (711)	98	2.0	100	0.0	NS
Old River, western arm at railroad bridge (902)	98	2.0	100	0.0	NS
Old River at mouth of Holland Cut (915)	100	0.0	94	6.1	NS
San Joaquin River between Hog and Turner Cuts (910)	98	1.8	100	0.0	NS
Sacramento Deep Water Channel, Light 55	98	2.0	100	0.0	NS
Suisun Bay off Chipps Island (508)	100	0.0	100	0.0	NS
Napa River at Vallejo Seawall (340)	100	0.0	100	0.0	NS
San Pablo Bay at Rodeo Flats (323)	79	6.9	83	3.8	NS
Montezuma Slough at Nurse Slough (609)	100	0.0	97	3.3	NS
Field Dup.: Old River at mouth of Holland Cut (915)	100	0.0	-	-	NA
Field Dup.: Sacramento River at tip of Grand Island (711)	94	2.5	-	-	NA

	MSD	PMSD
One-way ANOVA ³	-	-
Two-way ANOVA	15.0	15.3

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.085	0.008	0.108	0.019	NS
High EC Control @ 21.9 mS/cm	0.062	0.003	0.065	0.001	NS
Sacramento River at tip of Grand Island (711)	0.103	0.007	0.102	0.020	NS
Old River, western arm at railroad bridge (902)	0.116	0.008	0.103	0.011	NS
Old River at mouth of Holland Cut (915)	0.127	0.018	0.123	0.011	NS
San Joaquin River between Hog and Turner Cuts (910)	0.120	0.005	0.129	0.013	NS
Sacramento Deep Water Channel, Light 55	0.114	0.006	0.108	0.011	NS
Suisun Bay off Chipps Island (508)	0.128	0.015	0.138	0.008	NS
Napa River at Vallejo Seawall (340)	0.108	0.009	0.104	0.020	NS
San Pablo Bay at Rodeo Flats (323)	0.065	0.006	0.058	0.007	NS
Montezuma Slough at Nurse Slough (609)	0.117	0.015	0.114	0.012	NS
Field Dup.: Old River at mouth of Holland Cut (915)	0.100	0.012	-	-	NA
Field Dup.: Sacramento River at tip of Grand Island (711)	0.086	0.008	-	-	NA

	MSD	PMSD
One-way ANOVA	0.056	66.3
Two-way ANOVA	0.060	70.6

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. Survival in the unmanipulated samples was compared to the control using a Kruskal-Wallis test, and calculation of an MSD was not possible.

Table A25-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 6/27/06 - 6/28/06.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Sacramento River at tip of Grand Island (711)	130	22.6	7.81	7.8	8.4	0.23	0.007
Old River, western arm at railroad bridge (902)	155	24.7	7.61	7.5	5.6	0.04	0.001
Old River at mouth of Holland Cut (915)	158	25.5	7.42	7.7	7.9	0.03	0.000
San Joaquin River between Hog and Turner Cuts (910)	149	22.7	7.48	7.5	12.2	0.08	0.001
Sacramento Deep Water Channel, Light 55	367	24.3	7.28	7.0	23.2	0.02	0.000
Suisun Bay off Chipps Island (508)	269	21.7	7.66	7.8	21.0	0.00	0.000
Napa River at Vallejo Seawall (340)	11750	20.6	7.39	6.6	89.5	0.08	0.001
San Pablo Bay at Rodeo Flats (323)	20570	19.3	7.58	7.3	75.9	0.08	0.001
Montezuma Slough at Nurse Slough (609)	565	22.8	7.4	6.0	64.7	0.05	0.001
Field Dup.: Old River at mouth of Holland Cut (915)	158	25.5	7.42	7.7	9.1	0.03	0.000
Field Dup.: Sacramento River at tip of Grand Island (711)	130	22.6	7.81	7.8	7.2	0.19	0.005

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Table A25-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 6/29/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 6/27/06 - 6/28/06.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	374	24.9	25.5	6.0	8.3	7.49	9.27	108	55	-
High EC Control @ 21.9 mS/cm	21240	24.9	25.5	6.3	8.1	7.58	8.68	-	-	-
Sacramento River at tip of Grand Island (711)	165	24.5	25.4	6.0	8.7	7.65	9.35	52	52	0.022
Old River, western arm at railroad bridge (902)	171	24.7	25.5	6.0	8.8	7.50	8.58	46	34	0.003
Old River at mouth of Holland Cut (915)	186	24.5	25.4	6.2	8.6	7.48	8.32	36	37	0.001
San Joaquin River between Hog and Turner Cuts (910)	174	24.4	25.4	6.1	8.7	7.41	8.08	38	30	0.003
Sacramento Deep Water Channel, Light 55	382	25.0	25.5	5.7	8.8	7.75	8.45	108	92	0.001
Suisun Bay off Chipps Island (508)	274	24.7	25.4	6.1	8.4	7.55	8.46	60	47	0.000
Napa River at Vallejo Seawall (340)	11525	24.8	25.4	6.2	8.4	7.55	8.21	1360	70	0.002
San Pablo Bay at Rodeo Flats (323)	20860	24.9	25.5	6.3	8.8	7.56	8.08	2460	75	0.001
Montezuma Slough at Nurse Slough (609)	579	24.7	25.4	5.5	8.5	7.60	8.48	88	64	0.003
Field Dup.: Old River at mouth of Holland Cut (915)	186	24.7	25.5	6.2	8.5	7.54	8.76	46	36	0.001
Field Dup.: Sacramento River at tip of Grand Island (711)	161	24.7	25.5	6.0	8.7	7.61	8.43	52	53	0.010
DIEPAMHR + 100 ppb PBO	371	25.0	25.5	5.9	8.5	7.63	8.56	-	-	-
High EC Control @ 21.9 mS/cm + 100 ppb PBO	21505	24.8	25.5	7.0	8.4	7.72	8.30	-	-	-
Sacramento River at tip of Grand Island (711) + 100 ppb PBO	160	25.0	25.4	6.2	8.6	7.66	8.67	-	-	-
Old River, western arm at railroad bridge (902) + 100 ppb PBO	172	24.8	25.4	6.2	8.6	7.65	8.38	-	-	-
Old River at mouth of Holland Cut (915) + 100 ppb PBO	182	24.7	25.4	6.3	8.7	7.56	8.22	-	-	-
San Joaquin River between Hog and Turner Cuts (910) + 100 ppb PBO	171	24.6	25.4	6.1	8.8	7.39	8.28	-	-	-
Sacramento Deep Water Channel, Light 55 + 100 ppb PBO	374	24.7	25.4	6.1	8.6	7.82	8.59	-	-	-
Suisun Bay off Chipps Island (508) + 100 ppb PBO	267.2	24.7	25.4	6.4	8.7	7.64	8.42	-	-	-
Napa River at Vallejo Seawall (340) + 100 ppb PBO	11390	24.6	25.4	6.5	8.5	7.64	8.17	-	-	-
San Pablo Bay at Rodeo Flats (323) + 100 ppb PBO	20660	24.7	25.4	6.3	8.6	7.57	8.17	-	-	-
Montezuma Slough at Nurse Slough (609) + 100 ppb PBO	566	24.5	25.3	6.0	8.6	7.66	8.53	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A26-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 6/30/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 6/28/06 - 6/29/06.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	92	5.8	93	6.7	NS
Carquinez Strait, West of Benicia army dock (405)	98	2.0	100	0.0	NS
Grizzly Bay at Dolphin (602)	98	2.0	97	3.3	NS
Suisun Bay, East of middle point (504)	96	4.4	97	3.3	NS
Sacramento R. across from Sherman Lake (704)	92	5.4	90	0.4	NS
Middle of Broad Slough, West end (804)	90	3.2	90	5.8	NS
San Joaquin River, West of Oulton Point (812)	94	4.0	83	8.8	NS

	MSD	PMSD
One-way ANOVA ³	-	-
Two-way ANOVA	22.7	24.7

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.054	0.009	0.167	0.018	S (309%)
Carquinez Strait, West of Benicia army dock (405)	0.064	0.010	0.179	0.006	S (280%)
Grizzly Bay at Dolphin (602)	0.053	0.010	0.121	0.026	NS
Suisun Bay, East of middle point (504)	0.067	0.009	0.143	0.020	NS
Sacramento R. across from Sherman Lake (704)	0.038	0.010	0.074	0.009	NS
Middle of Broad Slough, West end (804)	0.134	0.035	0.145	0.038	NS
San Joaquin River, West of Oulton Point (812)	0.187	0.008	0.033	0.011	S (18%)

	MSD	PMSD
One-way ANOVA ³	-	-
Two-way ANOVA	0.090	165.9

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. Survival and weight in the unmanipulated samples were compared to the control using Kruskal-Wallis tests, and calculations of MSDs were not possible.

Table A26-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 6/28/06 - 6/29/06.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Carquinez Strait, West of Benicia army dock (405)	2924	21.1	7.69	7.6	205.7	0.10	0.002
Grizzly Bay at Dolphin (602)	662	20.8	7.41	8.6	120.7	0.02	0.000
Suisun Bay, East of middle point (504)	238	20.1	7.39	8.1	27.6	0.04	0.000
Sacramento R. across from Sherman Lake (704)	142	21.8	7.21	7.3	20.2	0.05	0.000
Middle of Broad Slough, West end (804)	138	22.6	7.61	7.8	8.7	0.00	0.000
San Joaquin River, West of Oulton Point (812)	133	22.9	7.42	7.8	9.7	0.00	0.000

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Table A26-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 6/30/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 6/28/06 - 6/29/06.

Treatment	Laboratory Chemistry						Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹	
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH				Max pH
DIEPAMHR	369	23.6	25.5	6.1	8.4	7.55	8.03	108	55	-
Carquinez Strait, West of Benicia army dock (405)	2478	23.5	25.5	6.2	8.1	7.36	7.85	308	55	0.003
Grizzly Bay at Dolphin (602)	665	23.7	25.4	6.1	8.8	7.50	7.97	96	54	0.001
Suisun Bay, East of middle point (504)	258	23.8	25.5	5.5	8.7	7.24	8.01	56	50	0.002
Sacramento R. across from Sherman Lake (704)	166	24.0	24.0	5.7	8.6	7.42	7.94	50	52	0.002
Middle of Broad Slough, West end (804)	168	23.9	24.2	5.2	8.2	7.51	8.00	42	42	0.000
San Joaquin River, West of Oulton Point (812)	149	23.7	23.7	5.7	8.4	7.54	7.95	40	40	0.000
DIEPAMHR + 50 ppb PBO	372	24.2	24.8	6.4	7.9	7.71	8.05	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 50 ppb PBO	2584	24.7	24.9	6.2	8.1	7.51	7.87	-	-	-
Grizzly Bay at Dolphin (602) + 50 ppb PBO	675	24.7	25.5	6.1	8.1	7.55	8.00	-	-	-
Suisun Bay, East of middle point (504) + 50 ppb PBO	266	24.7	25.7	6.1	8.1	7.58	7.63	-	-	-
Sacramento R. across from Sherman Lake (704) + 50 ppb PBO	180	24.7	25.3	6.0	8.3	7.55	7.99	-	-	-
Middle of Broad Slough, West end (804) + 50 ppb PBO	171	24.6	25.6	5.5	8.4	7.50	7.83	-	-	-
San Joaquin River, West of Oulton Point (812) + 50 ppb PBO	166	24.2	24.6	5.4	8.6	7.47	7.88	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A27-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 07/13/2006 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 7/11/2006 - 7/12/2006.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	100	0.0	96	3.7	NS
High EC Control @ 27.50 mS/cm	88	3.7	-	-	NA
Old R. at mouth of Holland Cut (902)	100	0.0	91	5.2	NS
Old R., Western Arm at Railroad Bridge (915)	100	0.0	100	0.0	NS
San Joaquin, Light 21 (910)	100	0.0	100	0.0	NS
Sac.R., at tip of Grand Island (711)	100	0.0	97	3.3	NS
Deep Water Channel @ Light 55	96	2.4	100	0.0	NS
S.P.Bay @ Rodeo Flats (323) ³	34	5.1	42	6.0	NS
Napa River @ Vallejo Seawall (340) ³	77	6.9	83	3.3	NS
Carquinez Straight @ Benicia (405)	98	2.0	100	0.0	NS
Suisun Bay @ Chipps Island (508)	98	1.8	97	3.3	NS
Montezuma Slough @ Nurse Slough (609)	96	4.0	93	3.3	NS

	MSD	PMSD
One-way ANOVA ⁴	-	-
Two-way ANOVA	17.3	17.3

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.101	0.002	0.082	0.006	NS
High EC Control @ 27.50 mS/cm	0.028	0.004	-	-	NA
Old R. at mouth of Holland Cut (902)	0.182	0.014	0.143	0.009	NS
Old R., Western Arm at Railroad Bridge (915)	0.134	0.009	0.151	0.004	NS
San Joaquin, Light 21 (910)	0.199	0.013	0.168	0.015	NS
Sac.R., at tip of Grand Island (711)	0.159	0.007	0.141	0.017	NS
Deep Water Channel @ Light 55	0.182	0.013	0.140	0.013	NS
S.P.Bay @ Rodeo Flats (323) ³	0.132	0.013	0.037	0.010	S (28%)
Napa River @ Vallejo Seawall (340) ³	0.165	0.025	0.097	0.008	NS
Carquinez Straight @ Benicia (405)	0.146	0.007	0.156	0.002	NS
Suisun Bay @ Chipps Island (508)	0.156	0.009	0.146	0.003	NS
Montezuma Slough @ Nurse Slough (609)	0.175	0.011	0.204	0.0	NS

	MSD	PMSD
One-way ANOVA ⁴	-	-
Two-way ANOVA	0.049	48.7

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. This high conductivity sample was compared to the High EC control @ 27.50 mS/cm.

4. Survival and weight in the unmanipulated samples were compared to the control using Kruskal-Wallis tests, and calculations of MSDs were not possible.

Table A27-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 7/11/2006 - 7/12/2006.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Old R. at mouth of Holland Cut (902)	223	24.1	8.18	7.06	5.08	0.04	0.003
Old R., Western Arm at Railroad Bridge (915)	214	24.6	7.29	7.41	6.54	0.02	0.000
San Joaquin, Light 21 (910)	344	24.2	7.08	6.01	12.43	0.20	0.001
Sac.R., at tip of Grand Island (711)	250	22.4	7.52	8.07	9.24	0.09	0.001
Deep Water Channel @ Light 55	413	23.1	7.79	7.24	68.90	0.10	0.003
S.P.Bay @ Rodeo Flats (323)	26420	19.9	7.71	7.71	73.43	0.08	0.001
Napa River @ Vallejo Seawall (340)	18110	19.9	7.58	7.58	25.53	0.04	0.000
Carquinez Straight @ Benicia (405)	9640	20.1	9.64	9.64	82.70	0.08	0.045
Suisun Bay @ Chipps Island (508)	987	22.1	7.57	8.10	15.80	0.00	0.000
Montezuma Slough @ Nurse Slough (609)	714	22.2	7.44	7.60	82.20	0.04	0.000

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Table A27-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 7/13/2006 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 7/11/2006 - 7/12/2006.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	378	24.8	25.8	5.3	8.2	7.62	7.92	108	63	-
High EC Control @ 27.50 mS/cm	26670	24.9	25.7	5.8	8.4	7.59	7.96	-	-	-
Old R. at mouth of Holland Cut (902)	189	24.9	25.6	5.4	8.6	7.58	8.10	46	44	0.002
Old R., Western Arm at Railroad Bridge (915)	194	24.8	25.5	5.2	8.2	7.54	8.22	42	40	0.002
San Joaquin, Light 21 (910)	325	24.7	25.6	5.9	8.3	7.58	7.79	72	52	0.006
Sac.R., at tip of Grand Island (711)	175	25.0	25.4	5.7	8.4	7.66	7.94	52	56	0.004
Deep Water Channel @ Light 55	389	24.7	25.6	5.9	8.3	7.86	8.09	108	88	0.006
S.P.Bay @ Rodeo Flats (323)	25700	24.4	25.5	5.6	8.3	7.55	7.62	2800	200	0.001
Napa River @ Vallejo Seawall (340)	17890	24.6	25.5	6.2	8.5	7.60	7.78	1960	160	0.001
Carquinez Straight @ Benicia (405)	9650	24.5	25.5	5.9	8.3	7.48	7.76	1040	120	0.002
Suisun Bay @ Chipps Island (508)	978	24.4	25.4	6.0	8.2	7.64	7.99	122	52	0.000
Montezuma Slough @ Nurse Slough (609)	744	24.6	25.4	6.1	8.2	7.72	8.18	102	70	0.003
DIEPAMHR + 50 ppb PBO	389	24.7	25.4	6.1	8.3	7.70	8.09	-	-	-
Old R. at mouth of Holland Cut (902) + 50 ppb PBO	184	24.5	25.4	6.1	8.4	7.60	8.22	-	-	-
Old R., Western Arm at Railroad Bridge (915) + 50 ppb PBO	193	24.6	25.4	6.0	8.3	7.58	7.93	-	-	-
San Joaquin, Light (910) + 50 ppb PBO	320	24.3	25.3	6.2	8.2	7.64	7.86	-	-	-
Sac.R., at tip of Grand Island (711) + 50 ppb PBO	191	24.4	25.3	6.3	8.4	7.72	8.09	-	-	-
Deep Water Channel @ Light 55 + 50 ppb PBO	383	24.3	25.3	6.4	8.4	7.86	8.24	-	-	-
S.P.Bay @ Rodeo Flats (323) + 50 ppb PBO	25795	24.3	25.2	6.0	8.3	7.59	7.69	-	-	-
Napa River @ Vallejo Seawall (340) + 50 ppb PBO	17590	24.2	25.3	6.5	8.3	7.64	7.80	-	-	-
Carquinez Straight @ Benicia (405) + 50 ppb PBO	9600	24.3	25.2	6.0	8.4	7.53	7.70	-	-	-
Suisun Bay @ Chipps Island (508) + 50 ppb PBO	982.5	24.1	25.3	6.0	8.4	7.62	8.14	-	-	-
Montezuma Slough @ Nurse Slough (609) + 50 ppb PBO	749.5	24.3	25.3	6.2	8.3	7.73	8.20	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A28-1. Survival of *H. azteca* in a Toxicity Identification Evaluation initiated on 7/24/06 examining an ambient water column sample collected at San Pablo Bay at Rodeo Flats (POD site 323) on 7/12/06.¹

Treatment	Survival (%) ²									
	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Day 8	Day 9	Day 10
DIEPAMHR	100	100	100	100	100	100	100	100	100	100
DIEPAMHR (HA) @ 1436 mg/L	100	100	100	100	100	100	100	100	100	100
DIEPAMHR (HA) + MeOH @ 0.5%	100	100	100	100	100	100	100	100	100	100
DIEPAMHR (HA) + eluate addback @ 3x	100	93	87	53	53	53	53	53	53	53
DIEPAMHR (HA) + 1436 mg/L EDTA	47	0	0	0	-	-	-	-	-	-
DIEPAMHR (HA) + 718 mg/L EDTA	92	68	30	8	-	-	-	-	-	-
DIEPAMHR (HA) + 359 mg/L EDTA	100	100	100	100	-	-	-	-	-	-
DIEPAMHR (HA) + 179.5 mg/L EDTA	100	100	100	100	-	-	-	-	-	-
DIEPAMHR (HA) + 2872 mg/L STS	100	80	49	11	-	-	-	-	-	-
DIEPAMHR (HA) + 1436 mg/L STS	100	82	66	66	-	-	-	-	-	-
DIEPAMHR (HA) + 718 mg/L STS	100	60	47	33	-	-	-	-	-	-
DIEPAMHR (HA) + 359 mg/L STS	100	92	92	28	-	-	-	-	-	-
DIEPAMHR (HA) air stripped	100	100	100	100	100	100	100	100	93	93
DIEPAMHR C8 Blank	100	100	100	100	100	100	100	100	100	100
323	100	100	100	100	100	100	100	92	83	83
323 + 2800 mg/L EDTA	100	0	0	0	-	-	-	-	-	-
323 + 1400 mg/L EDTA	100	20	0	0	-	-	-	-	-	-
323 + 700 mg/L EDTA	93	53	13	0	-	-	-	-	-	-
323 + 350 mg/L EDTA	100	80	80	53	-	-	-	-	-	-
323 + 5600 mg/L STS	100	20	0	0	-	-	-	-	-	-
323 + 2800 mg/L STS	100	7	0	0	-	-	-	-	-	-
323 + 1400 mg/L STS	100	13	13	0	-	-	-	-	-	-
323 + 700 mg/L STS	100	27	0	0	-	-	-	-	-	-
323 air stripped	100	100	100	100	67	67	67	67	53	53
323 C8 Rinsate	100	100	87	58	52	52	52	52	28	22

1. Sample was filtered through a C8 column on 7/21/06.
2. Highlighted areas indicate mean survival less than 50%.
3. EDTA and STS treated samples were only tested through Day 4, because they clearly reduced *Hyalella* survival relative to the untreated sample, indicating that metals were not detected as a cause of toxicity.

Table A29-1. Survival of *H. azteca* in a Toxicity Identification Evaluation initiated on 08/01/06 examining an ambient water column sample collected at San Pablo Bay at Rodeo Flats (POD site 323) on 7/12/06.¹

Treatment	Survival (%) ²									
	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Day 8	Day 9	Day 10
DIEPAMHR	100	100	100	100	100	100	100	100	100	100
Diluted Seawater @ 2740 mg/L	100	93	93	87	87	87	87	87	87	87
Diluted Seawater + 120 mg/L EDTA	100	100	93	80	73	73	73	73	73	73
Diluted Seawater + 60 mg/L EDTA	100	100	100	100	93	93	93	93	93	93
Diluted Seawater + 30 mg/L EDTA	100	100	100	100	89	89	82	82	82	82
Diluted Seawater + 25 mg/L STS	100	100	100	80	67	67	67	53	53	53
Diluted Seawater + 12.5 mg/L STS	93	93	87	80	80	80	73	73	73	73
Diluted Seawater + 6.25 mg/L STS	100	100	93	80	73	73	67	67	67	67
POD 323	100	92	92	92	92	83	83	83	83	83
POD 323 + 120 mg/L EDTA	100	100	100	87	87	87	87	87	87	87
POD 323 + 60 mg/L EDTA	87	87	80	73	73	73	73	73	73	73
POD 323 + 30 mg/L EDTA	93	93	65	50	50	50	43	43	37	37
POD 323 + 25 mg/L STS	93	93	37	37	37	37	37	37	28	28
POD 323 + 12.5 mg/L STS	100	100	93	80	80	73	73	73	73	73
POD 323 + 6.25 mg/L STS	100	100	93	67	67	60	60	60	60	60
Diluted Seawater @ 20 ppt	20	20	13	7	7	0	0	0	0	0
Diluted Seawater @ 18 ppt	60	53	27	20	20	20	13	13	13	13
Diluted Seawater @ 16 ppt	80	80	73	67	67	53	53	53	53	53
Diluted Seawater @ 14 ppt	93	93	93	87	87	80	80	80	80	80
Diluted Seawater @ 12 ppt	93	93	87	87	87	87	87	87	87	87

1. Sample was filtered through a C8 column on 7/21/06.
2. Highlighted areas indicate mean survival less than 50%.

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Table A30-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 07/14/2006 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 7/13/2006.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	98	2.0	100	0.0	NS
Grizzly Bay @ Dolphin (602)	98	2.0	100	0.0	NS
Suisun Bay @ Middle Point (504)	84	9.3	97	3.0	NS
Sac. River @ Sherman Lake (704)	100	0.0	97	3.3	NS
Broad Slough, West End (804)	98	2.0	100	0.0	NS
SJR @ West of Oulton Point (812)	100	0.0	100	0.0	NS

	MSD	PMSD
One-way ANOVA ³	-	-
Two-way ANOVA	18.8	19.2

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.114	0.006	0.145	0.007	NS
Grizzly Bay @ Dolphin (602)	0.159	0.007	0.166	0.014	NS
Suisun Bay @ Middle Point (504)	0.169	0.021	0.148	0.008	NS
Sac. River @ Sherman Lake (704)	0.192	0.013	0.178	0.010	NS
Broad Slough, West End (804)	0.177	0.005	0.172	0.000	NS
SJR @ West of Oulton Point (812)	0.169	0.004	0.173	0.003	NS

	MSD	PMSD
One-way ANOVA ³	-	-
Two-way ANOVA	0.054	46.9

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. Survival and weight in the unmanipulated samples were compared to the control using Kruskal-Wallis tests, and calculations of MSDs were not possible.

Table A30-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 7/13/2006.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Grizzly Bay @ Dolphin (602)	5800	21.1	7.85	8.34	62.10	0.05	0.001
Suisun Bay @ Middle Point (504)	6700	21.3	7.49	7.90	13.87	0.04	0.001
Sac. River @ Sherman Lake (704)	225.4	22.2	7.73	8.10	24.40	0.04	0.001
Broad Slough, West End (804)	229.3	23.6	7.35	7.50	9.41	0.04	0.000
SJR @ West of Oulton Point (812)	145.7	23.3	7.50	7.70	10.27	0.02	0.000

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Table A30-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 07/14/2006 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 7/13/2006.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	364	24.5	24.8	5.3	8.2	7.46	8.03	108	63	-
Grizzly Bay @ Dolphin (602)	5370	24.0	24.4	5.3	8.3	7.23	7.77	610	80	0.0012
Suisun Bay @ Middle Point (504)	6615	24.4	24.6	4.9	8.8	7.21	7.64	798	91	0.0007
Sac. River @ Sherman Lake (704)	256	24.7	24.7	5.0	8.6	7.32	8.04	56	55	0.0022
Broad Slough, West End (804)	254	24.5	24.9	5.1	8.9	7.40	7.99	56	48	0.0019
SJR @ West of Oulton Point (812)	172	24.4	26.1	5.6	8.6	7.41	7.90	48	49	0.0008
DIEPAMHR + 50 ppb PBO	375	24.3	26.1	5.4	8.3	7.37	8.09	-	-	-
Grizzly Bay @ Dolphin (602) + 50 ppb PBO	5385	24.5	26.3	5.4	8.7	7.24	7.82	-	-	-
Suisun Bay @ Middle Point (504) + 50 ppb PBO	6570	24.8	26.2	5.1	8.6	7.21	7.69	-	-	-
Sac. River @ Sherman Lake (704) + 50 ppb PBO	289	24.6	26.3	5.5	8.6	7.43	8.01	-	-	-
Broad Slough, West End (804) + 50 ppb PBO	254	24.7	26.1	5.1	8.2	7.24	7.97	-	-	-
SJR @ West of Oulton Point (812) + 50 ppb PBO	172	24.7	26.1	5.0	8.3	7.30	7.95	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A31-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 07/27/2006 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 07/25/2006 and 07/26/2006.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
Lab Control (DIEPAMHR)	98	2.0	97	3.3	NS
Sacramento River Deep Water Channel, Light 55	100	0.0	100	0.0	NS
Sacramento River at tip of Grand Island (711)	100	0.0	100	0.0	NS
Old River - Western Arm to Railroad Bridge (902)	100	0.0	100	0.0	NS
San Joaquin River between Hog and Turner Cut (910)	98	2.2	100	0.0	NS
Old River at Holland Cut (915)	98	1.8	93	3.3	NS
Suisun Bay at Chipps Island (508)	90	2.9	100	0.0	NS
Montezuma Slough at Nurse Slough (609)	100	0.0	97	3.3	NS
Broad Slough, West End (804)	100	0.0	100	0.0	NS
Sacramento River, North side across from Sherman Lake (704)	100	0.0	96	3.7	NS
San Joaquin River, West of Oulton Point (812)	96	2.4	100	0.0	NS
Field Duplicate: Old River at Holland Cut (915)	100	0.0	97	3.3	NS

	MSD	PMSD
One-way ANOVA ³	-	-
Two-way ANOVA	9.4	9.5

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
Lab Control (DIEPAMHR)	0.066	0.005	0.060	0.002	NS
Sacramento River Deep Water Channel, Light 55	0.101	0.005	0.086	0.003	NS
Sacramento River at tip of Grand Island (711)	0.078	0.004	0.070	0.008	NS
Old River - Western Arm to Railroad Bridge (902)	0.084	0.010	0.050	0.004	NS
San Joaquin River between Hog and Turner Cut (910)	0.096	0.008	0.086	0.003	NS
Old River at Holland Cut (915)	0.066	0.002	0.068	0.015	NS
Suisun Bay at Chipps Island (508)	0.070	0.006	0.073	0.010	NS
Montezuma Slough at Nurse Slough (609)	0.103	0.019	0.088	0.015	NS
Broad Slough, West End (804)	0.100	0.007	0.088	0.011	NS
Sacramento River, North side across from Sherman Lake (704)	0.077	0.007	0.077	0.004	NS
San Joaquin River, West of Oulton Point (812)	0.071	0.005	0.084	0.002	NS
Field Duplicate: Old River at Holland Cut (915)	0.086	0.006	0.082	0.003	NS

	MSD	PMSD
One-way ANOVA ³	-	-
Two-way ANOVA	0.046	69.1

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. Survival and weight in the unmanipulated samples were compared to the control using Kruskal-Wallis tests, and calculations of MSDs were not possible.

Table A31-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 07/25/2006 - 07/26/2006.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC ¹ (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Sacramento River Deep Water Channel, Light 55	-	28.6	7.89	6.97	13.13	0.00	-
Sacramento River at tip of Grand Island (711)	-	25.0	7.57	7.37	4.73	0.08	-
Old River - Western Arm to Railroad Bridge (902)	-	27.2	7.96	7.61	3.32	0.00	-
San Joaquin River between Hog and Turner Cut (910)	-	28.6	7.19	6.83	5.45	0.07	-
Old River at Holland Cut (915)	-	28.0	7.52	6.35	3.81	0.07	-
Suisun Bay at Chipps Island (508)	6810	24.7	7.78	7.26	10.77	0.00	0.000
Montezuma Slough at Nurse Slough (609)	2323	26.3	6.76	5.13	39.53	0.04	0.000
Broad Slough, West End (804)	263	26.5	6.98	6.54	4.77	0.07	0.000
Sacramento River, North side across from Sherman Lake (704)	279	25.3	7.24	6.85	14.30	0.05	0.000
San Joaquin River, West of Oulton Point (812)	178	26.3	6.96	6.50	8.17	0.07	0.000
Field Duplicate: Old River at Holland Cut (915)	-	28.0	7.52	6.35	3.42	0.10	-

1: Instrument malfunctioned at some sites.

Table A31-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 07/27/2006 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 07/25/2006 - 07/26/2006.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
Lab Control (DIEPAMHR)	359	24.1	26.1	6.5	8.3	7.62	8.09	108	59	-
Sacramento River Deep Water Channel, Light 55	318	24.6	26.0	6.1	8.3	7.70	8.14	96	80	0.000
Sacramento River at tip of Grand Island (711)	146	22.6	24.3	5.7	8.3	7.59	8.07	52	52	0.004
Old River - Western Arm to Railroad Bridge (902)	176	24.4	24.5	5.8	8.2	7.63	8.05	60	38	0.000
San Joaquin River between Hog and Turner Cut (910)	384	24.3	24.4	6.3	8.2	7.59	8.07	92	66	0.004
Old River at Holland Cut (915)	170	24.3	24.6	5.7	8.2	7.66	8.04	52	50	0.004
Suisun Bay at Chipps Island (508)	168	24.3	25.9	5.8	8.3	7.43	8.01	56	58	0.000
Montezuma Slough at Nurse Slough (609)	6200	24.2	26.0	5.9	8.3	7.30	7.78	744	70	0.001
Broad Slough, West End (804)	2223	24.2	24.8	6.4	8.2	7.30	7.79	280	66	0.002
Sacramento River, North side across from Sherman Lake (704)	277	24.2	25.2	5.9	8.2	7.54	8.01	68	52	0.002
San Joaquin River, West of Oulton Point (812)	292	24.2	26.1	6.1	8.2	7.48	8.06	68	56	0.004
Field Duplicate: Old River at Holland Cut (915)	188	24.3	26.2	6.0	8.2	7.43	8.08	60	52	0.006
Lab Control (DIEPAMHR) + 25 ppb PBO	352	23.7	25.1	6.3	8.3	7.65	8.12	-	-	-
Sacramento River Deep Water Channel, Light 55 + 25 ppb	238	23.9	25.1	6.3	8.3	7.57	8.21	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	163	23.9	25.3	6.1	8.4	7.58	8.18	-	-	-
Old River - Western Arm to Railroad Bridge (902) + 25 ppb PBO	173	23.9	25.5	6.2	8.2	7.46	8.04	-	-	-
San Joaquin River between Hog and Turner Cut (910) + 25 ppb PBO	375	23.8	25.4	6.3	8.4	7.61	8.05	-	-	-
Old River at Holland Cut (915) + 25 ppb PBO	186	23.8	25.9	6.1	8.3	7.46	8.07	-	-	-
Suisun Bay at Chipps Island (508) + 25 ppb PBO	174	23.8	26.4	6.2	8.3	7.46	8.05	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	6120	23.9	26.2	6.0	8.2	7.37	7.94	-	-	-
Broad Slough, West End (804) + 25 ppb PBO	2182	24.1	26.2	6.5	8.2	7.64	8.10	-	-	-
Sacramento River, North side across from Sherman Lake (704) + 25 ppb PBO	1636	23.9	26.3	6.3	8.3	7.54	8.11	-	-	-
San Joaquin River, West of Oulton Point (812) + 25 ppb PBO	282	24.1	26.5	6.3	8.5	7.63	8.10	-	-	-
Field Duplicate: Old River at Holland Cut (915) + 25 ppb PBO	178	24.0	26.5	6.2	8.3	7.56	8.05	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A32-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 07/28/2006 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 07/27/2006.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO ²
DIEPAMHR	100	0.0	90	5.8	NS
High EC Control @ 30.00 mS	0	0.0	0	0.0	NS
Grizzly Bay @ Dolphin(602) ³	93	6.7	97	3.0	NS
Napa River @ Vallejo Seawall (340) ³	46	14.9	31	20.7	NS
S.P. Bay @ Rodeo Flats (323) ³	5	2.8	15	9.8	NS
Carquinez Straight @ Benicia (405) ³	91	6.5	70	9.8	NS
Suisun Bay @ Middle Point (504)	94	2.4	97	3.3	NS
Bottle Blank	82	7.2	-	-	NA

	MSD	PMSD
One-way ANOVA ⁴	-	-
Two-way ANOVA	44.9	44.9

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO ²
DIEPAMHR	0.056	0.004	0.044	0.003	NS
High EC Control @ 30.00 mS	-	-	-	-	NA
Grizzly Bay @ Dolphin(602) ³	0.049	0.009	0.056	0.015	NS
Napa River @ Vallejo Seawall (340) ³	0.031	0.008	0.007	0.015	NS
S.P. Bay @ Rodeo Flats (323) ³	0.010	0.010	0.003	0.009	NS
Carquinez Straight @ Benicia (405) ³	0.059	0.002	0.036	0.005	NS
Suisun Bay @ Middle Point (504)	0.077	0.010	0.053	0.011	NS
Bottle Blank	0.035	0.005	-	-	NA

	MSD	PMSD
One-way ANOVA ⁴	-	-
Two-way ANOVA	0.042	75.5

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. Survival and weight in these high conductivity samples could not be evaluated statistically due to 100% mortality in the High EC Control @ 30.00 mS/cm.

4. Survival and weight in the unmanipulated samples were compared to the control using Kruskal-Wallis tests, and calculations of MSDs were not possible.

Table A32-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 07/27/2006.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Grizzly Bay @ Dolphin (602)	11500	22.6	7.94	7.65	200.67	0.12	0.003
Napa River @ Vallejo Seawall (340)	23560	22.2	7.32	6.16	15.20	0.04	0.000
S.P. Bay @ Rodeo Flats (323)	30260	21.9	7.32	6.73	19.77	0.08	0.000
Carquinez Straight @ Benicia (405)	15520	23.1	7.29	6.95	97.40	0.13	0.001
Suisun Bay @ Middle Point (504)	5140	24.6	7.14	6.98	19.97	0.05	0.000
Bottle Blank	289	19.9	8.4	8.70	0.50	0.01	0.001

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Table A32-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 07/28/2006 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 07/27/2006.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	354	22.7	24.2	6.1	8.1	7.60	8.04	108	63	-
High EC Control @ 30.00 mS	30575	22.1	24.2	8.1	8.7	7.86	7.88	-	-	-
Grizzly Bay @ Dolphin(602)	10865	22.8	24.2	6.4	8.3	7.38	7.85	1200	110	0.003
Napa River @ Vallejo Seawall (340)	21540	22.8	24.2	5.8	8.2	7.38	7.70	2400	250	0.001
S.P. Bay @ Rodeo Flats (323)	28295	22.8	24.0	6.1	8.4	7.46	7.73	3450	250	0.001
Carquinez Straight @ Benicia (405)	14460	22.8	24.0	6.5	8.2	7.50	7.76	3400	100	0.003
Suisun Bay @ Middle Point (504)	5070	22.8	24.0	6.4	8.4	7.55	7.81	500	30	0.001
Bottle Blank	393	22.8	24.2	6.3	8.2	7.62	8.16	68	60	0.001
DIEPAMHR + 25 ppb PBO	364	22.8	23.3	6.5	8.3	7.77	8.09	-	-	-
High EC Control @ 30.00 mS + 25 ppb PBO	28890	23.3	23.3	6.8	8.4	7.58	7.88	-	-	-
Grizzly Bay @ Dolphin(602) + 25 ppb PBO	10680	22.8	23.3	6.6	8.4	7.50	7.84	-	-	-
Napa River @ Vallejo Seawall (340) + 25 ppb PBO	21665	22.8	23.7	6.2	8.2	7.49	7.77	-	-	-
S.P. Bay @ Rodeo Flats (323) + 25 ppb PBO	26295	22.8	23.3	6.3	8.5	7.47	7.80	-	-	-
Carquinez Straight @ Benicia (405) + 25 ppb PBO	14235	22.8	23.3	6.2	8.4	7.52	7.81	-	-	-
Suisun Bay @ Middle Point (504) + 25 ppb PBO	5015	22.8	23.2	6.4	8.2	7.49	7.95	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A33-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 8/10/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 8/08/06 - 8/09/06.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO ²
DIEPAMHR	96	2.4	100	0.0	NS
High EC Control @ 13.85 mS/cm	90	3.2	89	6.4	NS
High EC Control @ 22.7 mS/cm	25	7.2	85	9.8	S (340%)
Sacramento Deep Water Channel, Light 55	98	1.8	100	0.0	NS
Sacramento River at tip of Grand Island (711)	98	2.0	97	3.3	NS
Old River, western arm at railroad bridge (902)	98	2.0	100	0.0	NS
San Joaquin River between Hog and Turner Cuts (910)	100	0.0	100	0.0	NS
Old River at mouth of Holland Cut (915)	96	2.4	100	0.0	NS
Suisun Bay off Chipps Island (508)	100	0.0	100	0.0	NS
Montezuma Slough at Nurse Slough (609)	100	0.0	100	0.0	NS
Napa River at Riverside Blvd. terminus (340)	86	4.2	82	7.8	NS
Carquinez Strait, West of Benicia army dock (405)	84	4.0	93	3.3	NS
Field Dup.: Carquinez Strait, West of Benicia army dock (405)	94	4.0	-	-	NA

	MSD	PMSD
One-way ANOVA	15.5	16.2
Two-way ANOVA	18.2	18.9

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO ²
DIEPAMHR	0.057	0.008	0.038	0.008	NS
High EC Control @ 13.85 mS/cm	0.067	0.006	0.055	0.007	NS
High EC Control @ 22.7 mS/cm	0.037	0.005	0.037	0.003	NS
Sacramento Deep Water Channel, Light 55	0.111	0.005	0.126	0.003	NS
Sacramento River at tip of Grand Island (711)	0.098	0.008	0.069	0.006	NS
Old River, western arm at railroad bridge (902)	0.094	0.007	0.099	0.007	NS
San Joaquin River between Hog and Turner Cuts (910)	0.140	0.008	0.125	0.014	NS
Old River at mouth of Holland Cut (915)	0.071	0.019	0.087	0.018	NS
Suisun Bay off Chipps Island (508)	0.106	0.016	0.107	0.011	NS
Montezuma Slough at Nurse Slough (609)	0.103	0.018	0.153	0.016	NS
Napa River at Riverside Blvd. terminus (340)	0.094	0.033	0.043	0.002	NS
Carquinez Strait, West of Benicia army dock (405)	0.078	0.004	0.083	0.005	NS
Field Dup.: Carquinez Strait, West of Benicia army dock (405)	0.074	0.006	-	-	NA

	MSD	PMSD
One-way ANOVA	0.066	116.0
Two-way ANOVA	0.072	125.3

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

Table A33-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 8/08/06 - 8/09/06.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Sacramento Deep Water Channel, Light 55	274	22.0	7.95	7.9	13.7	0.05	0.002
Sacramento River at tip of Grand Island (711)	145	21.1	7.68	8.1	7.7	0.08	0.002
Old River, western arm at railroad bridge (902)	230	22.5	8.45	7.6	4.1	0.02	0.002
San Joaquin River between Hog and Turner Cuts (910)	437	24.9	7.41	5.3	12.9	0.44	0.006
Old River at mouth of Holland Cut (915)	234	24.2	7.58	7.0	4.9	0.06	0.001
Suisun Bay off Chipps Island (508)	4526	21.3	7.62	8.2	11.5	0.08	0.001
Montezuma Slough at Nurse Slough (609)	4200	22.4	7.58	7.8	35.5	0.05	0.001
Napa River at Riverside Blvd. terminus (340)	22700	22.1	7.84	6.3	16.0	0.00	0.000
Carquinez Strait, West of Benicia army dock (405)	13850	22.2	7.84	7.6	45.0	0.05	0.001
Field Dup.: Carquinez Strait, West of Benicia army dock (405)	13850	22.2	7.84	7.6	45.0	0.00	0.000

Table A33-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 8/10/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 8/08/06 and 8/09/06.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	335	24.8	25.1	5.8	8.3	7.55	8.10	108	63	-
High EC Control @ 13.85 mS	13560	24.9	24.9	6.6	8.4	7.59	7.82	1560	100	-
High EC Control @ 22.7 mS	22285	24.7	25.0	6.6	8.4	7.55	7.79	2400	180	-
Sacramento Deep Water Channel, Light 55	288	24.8	24.9	5.9	8.3	7.72	8.26	80	76	0.004
Sacramento River at tip of Grand Island (711)	225	25.0	25.0	5.9	8.2	7.64	8.21	54	58	0.006
Old River, western arm at railroad bridge (902)	241	24.9	24.9	6.1	8.4	7.56	8.22	58	52	0.002
San Joaquin River between Hog and Turner Cuts (910)	436	22.8	24.5	5.8	8.5	7.58	7.91	100	71	0.015
Old River at mouth of Holland Cut (915)	239	24.3	24.6	5.9	8.4	7.55	8.00	64	56	0.003
Suisun Bay off Chipps Island (508)	2381	23.6	24.9	6.2	8.4	7.49	7.89	476	72	0.002
Montezuma Slough at Nurse Slough (609)	4023	24.1	24.8	6.2	8.3	7.57	7.89	452	82	0.002
Napa River at Riverside Blvd. terminus (340)	21530	24.4	24.7	5.6	8.2	7.41	7.69	3080	220	0.000
Carquinez Strait, West of Benicia army dock (405)	13005	24.2	24.7	6.4	8.2	7.48	7.80	1480	110	0.001
Field Dup.: Carquinez Strait, West of Benicia army dock (405)	6441	23.7	24.6	6.2	8.4	7.42	7.71	2860	100	0.000
DIEPAMHR + 25 ppb PBO	338	24.7	24.9	5.8	8.4	7.62	8.16	-	-	-
High EC Control @ 22.7 mS + 25 ppb PBO	22270	24.8	24.9	6.2	8.3	7.48	7.84	-	-	-
High EC Control @ 13.85 mS + 25 ppb PBO	13595	24.6	24.9	6.7	8.5	7.57	7.80	-	-	-
Sacramento Deep Water Channel, Light 55 + 25 ppb PBO	351	24.7	25.0	6.0	8.5	7.69	8.24	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	166	24.6	25.0	5.6	8.6	7.60	8.10	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	242	24.4	24.9	6.0	8.6	7.56	8.11	-	-	-
San Joaquin River between Hog and Turner Cuts (910) + 25 ppb PBO	432	23.8	24.8	6.1	8.6	7.49	7.94	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	240.85	23.5	25.0	6.0	8.6	7.51	7.95	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	4400	23.9	25.0	6.0	8.7	7.26	7.75	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	4032	24.3	25.0	6.1	8.7	7.39	7.83	-	-	-
Napa River at Riverside Blvd. terminus (340) + 25 ppb PBO	20835	24.0	25.0	6.0	8.2	7.60	7.65	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	12940	24.6	25.0	6.6	8.5	7.47	7.82	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A34-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 08/11/2006 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 8/10/2006.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	98	2.0	97	3.3	NS
High EC Control @ 10.38 mS/cm	100	0.0	87	3.3	NS
Suisun Bay, East of middle point (504)	94	2.4	100	0.0	NS
Grizzly Bay at Dolphin (602)	92	4.1	80	11.5	NS
Sacramento R. across from Sherman Lake (704)	98	2.0	94	3.2	NS
Middle of Broad Slough, West end (804)	100	0.0	93	3.3	NS
San Joaquin River, West of Oulton Point (812)	98	2.0	93	3.5	NS

	MSD	PMSD
One-way ANOVA ³	-	-
Two-way ANOVA	16.6	17.0

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.116	0.012	0.069	0.011	NS
High EC Control @ 10.38 mS/cm	0.100	0.004	0.080	0.008	NS
Suisun Bay, East of middle point (504)	0.110	0.010	0.089	0.009	NS
Grizzly Bay at Dolphin (602)	0.059	0.005	0.086	0.008	NS
Sacramento R. across from Sherman Lake (704)	0.135	0.020	0.135	0.007	NS
Middle of Broad Slough, West end (804)	0.171	0.006	0.127	0.003	NS
San Joaquin River, West of Oulton Point (812)	0.124	0.012	0.131	0.012	NS

	MSD	PMSD
One-way ANOVA	0.056	48.6
Two-way ANOVA	0.056	48.6

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. Survival in the unmanipulated samples was compared to the control using a Kruskal-Wallis test, and calculation of an MSD was not possible.

Table A34-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 08/10/2006.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Suisun Bay, East of middle point (504)	10230	21.5	7.84	8.0	31.8	0.06	0.001
Grizzly Bay at Dolphin (602)	10380	22.4	8.14	8.5	12.4	0.03	0.001
Sacramento R. across from Sherman Lake (704)	229	21.9	7.92	8.1	20.8	0.09	0.003
Middle of Broad Slough, West end (804)	340	22.8	8.01	8.0	11.1	0.03	0.001
San Joaquin River, West of Oulton Point (812)	206	24.3	7.88	7.7	8.4	0.05	0.002

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Table A34-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 08/11/2006 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 08/10/2006.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	309	24.2	26.3	6.1	8.5	7.59	8.10	108	63	-
High EC Control @ 10.38 mS/cm	10090	24.1	26.3	6.4	8.9	7.37	7.94	1200	120	-
Suisun Bay, East of middle point (504)	9685	24.1	26.2	6.4	8.5	7.40	7.81	980	190	0.002
Grizzly Bay at Dolphin (602)	9760	24.1	26.1	6.5	8.8	7.37	7.94	980	110	0.001
Sacramento R. across from Sherman Lake (704)	331	24.1	26.1	6.5	8.6	7.80	8.19	60	60	0.007
Middle of Broad Slough, West end (804)	334	24.1	26.1	6.1	8.5	7.66	8.23	68	36	0.003
San Joaquin River, West of Oulton Point (812)	207	24.3	26.0	6.1	8.4	7.64	7.97	56	58	0.003
DIEPAMHR + 25 ppb PBO	316	24.1	25.4	6.3	8.5	7.56	8.19	-	-	-
High EC Control @ 10.38 mS/cm + 25 ppb PBO	9935	24.1	25.4	6.7	8.8	7.22	7.96	-	-	-
Suisun Bay, East of middle point (504) + 25 ppb PBO	9585	24.1	25.2	6.8	8.4	7.33	7.86	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	5245	24.3	25.2	6.8	8.5	7.37	7.96	-	-	-
Sacramento R. across from Sherman Lake (704) + 25 ppb PBO	287	24.2	25.2	6.3	8.7	7.67	8.02	-	-	-
Middle of Broad Slough, West end (804) + 25 ppb PBO	336	24.1	25.1	6.0	8.5	7.63	8.13	-	-	-
San Joaquin River, West of Oulton Point (812) + 25 ppb PBO	186	24.1	25.0	6.2	8.8	7.54	8.06	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A35-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 08/24/2006 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 08/22/06 - 08/23/06.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	100	0.0	97	3.3	NS
High EC Control @ 19 mS/cm	65	10.3	44	23.1	NS
Old River at mouth of Holland Cut (915)	98	2.0	93	6.7	NS
San Joaquin River between Hog and Turner Cuts (910)	100	0.0	100	0.0	NS
Old River, western arm at railroad bridge (902)	96	2.4	100	0.0	NS
Sacramento River at tip of Grand Island (711)	98	2.0	43	26.0	S (44%)
Sacramento Deep Water Channel, Light 55	100	0.0	97	3.0	NS
Montezuma Slough at Nurse Slough (609)	98	2.0	100	0.0	NS
Suisun Bay off Chipps Island (508)	98	1.8	97	3.3	NS
Carquinez Strait, West of Benicia army dock (405) ³	86	5.1	77	6.7	NS
Grizzly Bay at Dolphin (602)	96	2.8	100	0.0	NS
Napa River at Vallejo Seawall (340) ³	92	3.7	86	3.0	NS
Field Duplicate: Montezuma Slough at Nurse Slough (609)	92	3.8	-	-	NA

	MSD	PMSD
One-way ANOVA	19.8	19.8
Two-way ANOVA	24.0	24.0

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.090	0.005	0.076	0.010	NS
High EC Control @ 19 mS/cm	0.036	0.001	0.033	0.001	NS
Old River at mouth of Holland Cut (915)	0.131	0.007	0.126	0.011	NS
San Joaquin River between Hog and Turner Cuts (910)	0.135	0.010	0.128	0.005	NS
Old River, western arm at railroad bridge (902)	0.124	0.016	0.059	0.007	S (48%)
Sacramento River at tip of Grand Island (711)	0.105	0.006	0.039	0.011	S (37%)
Sacramento Deep Water Channel, Light 55	0.138	0.004	0.065	0.014	S (47%)
Montezuma Slough at Nurse Slough (609)	0.106	0.010	0.048	0.006	S (45%)
Suisun Bay off Chipps Island (508)	0.100	0.016	0.064	0.009	NS
Carquinez Strait, West of Benicia army dock (405) ³	0.060	0.005	0.065	0.003	NS
Grizzly Bay at Dolphin (602)	0.105	0.012	0.070	0.014	NS
Napa River at Vallejo Seawall (340) ³	0.062	0.005	0.048	0.005	NS
Field Duplicate: Montezuma Slough at Nurse Slough (609)	0.134	0.005	-	-	NA

	MSD	PMSD
One-way ANOVA	0.052	58.3
Two-way ANOVA	0.052	57.5

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. This high conductivity sample was compared to the 19 mS/cm High EC control.

Table A35-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 08/22/2006 - 08/23/2006.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Old River at mouth of Holland Cut (915)	259	22.0	7.80	7.8	4.0	0.02	0.001
San Joaquin River between Hog and Turner Cuts (910)	409	22.3	7.19	6.1	10.3	0.26	0.002
Old River, western arm at railroad bridge (902)	284	21.5	8.02	8.3	4.1	0.01	0.000
Sacramento River at tip of Grand Island (711)	155	21.8	7.29	7.8	8.0	0.19	0.002
Sacramento Deep Water Channel, Light 55	356	23.9	7.86	7.9	23.4	0.04	0.001
Montezuma Slough at Nurse Slough (609)	5420	20.4	7.42	7.8	74.6	0.10	0.001
Suisun Bay off Chipps Island (508)	3699	19.9	7.45	8.1	23.2	0.08	0.001
Carquinez Strait, West of Benicia army dock (405)	1795	20.3	7.62	7.9	35.0	0.13	0.002
Grizzly Bay at Dolphin (602)	1086	19.5	7.63	8.1	101.5	0.07	0.001
Napa River at Vallejo Seawall (340)	1683	22.7	7.51	6.7	6.2	0.02	0.000
Field Duplicate: Montezuma Slough at Nurse Slough (609)	-	-	-	-	64.8	0.10	-

Table A35-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 08/24/2006 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 08/22/2006 - 08/23/2006.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	352	23.3	24.1	6.6	8.4	7.58	8.15	106	59	-
High EC Control @ 19 mS/cm	18945	23.2	24.1	6.8	8.3	7.49	8.09	3160	160	-
Old River at mouth of Holland Cut (915)	263	23.1	24.0	6.0	8.6	7.56	8.26	68	58	0.002
San Joaquin River between Hog and Turner Cuts (910)	400	23.1	24.2	6.1	8.5	7.62	8.08	100	74	0.015
Old River, western arm at railroad bridge (902)	283	23.0	24.2	6.1	8.4	7.49	8.35	68	58	0.001
Sacramento River at tip of Grand Island (711)	159	23.0	24.2	5.9	8.4	7.52	8.10	56	64	0.011
Sacramento Deep Water Channel, Light 55	342	23.0	24.3	5.9	8.6	7.67	8.23	104	90	0.003
Montezuma Slough at Nurse Slough (609)	5050	23.0	24.2	6.5	8.4	7.51	7.97	720	85	0.004
Suisun Bay off Chipps Island (508)	3533	22.9	24.1	6.7	8.5	7.52	7.95	396	68	0.003
Carquinez Strait, West of Benicia army dock (405)	17335	22.7	24.2	6.8	8.6	7.43	7.75	3600	180	0.003
Grizzly Bay at Dolphin (602)	9765	22.7	24.3	6.7	8.8	7.46	7.86	1120	120	0.002
Napa River at Vallejo Seawall (340)	15715	22.7	24.2	6.7	8.7	7.75	7.85	3720	260	0.000
Field Duplicate: Montezuma Slough at Nurse Slough (609)	5050	22.9	24.2	6.8	8.4	7.58	8.01	670	85	0.004
DIEPAMHR + 25 ppb PBO	353	22.7	23.7	6.6	8.4	7.62	8.23	-	-	-
High EC Control @ 19 mS/cm + 25 ppb PBO	18465	22.6	23.7	6.7	8.4	7.55	8.06	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	269	22.7	23.6	6.3	8.4	7.65	8.31	-	-	-
San Joaquin River between Hog and Turner Cuts (910) + 25 ppb PBO	484	22.8	23.6	6.4	8.4	7.68	8.21	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	286	22.6	23.7	6.3	8.6	7.58	8.34	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	166	22.5	23.6	6.6	8.5	7.69	8.16	-	-	-
Sacramento Deep Water Channel, Light 55 + 25 ppb PBO	345	22.6	23.6	6.1	8.5	7.69	8.27	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	2761.5	22.6	23.6	6.9	8.5	7.60	7.92	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	3460	22.4	23.6	6.5	8.5	7.50	8.01	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	17035	22.3	23.6	6.8	8.5	7.49	7.75	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	9585	22.1	23.5	6.9	8.3	7.49	7.93	-	-	-
Napa River at Vallejo Seawall (340) + 25 ppb PBO	15635	22.2	23.6	6.7	8.7	7.75	7.90	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A36-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 08/25/2006 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 08/23/2006 - 08/24/2006.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO ²
DIEPAMHR	100	0.0	100	0.0	NS
Suisun Bay, East of middle point (504)	100	0.0	100	0.0	NS
Middle of Broad Slough, West end (804)	98	2.0	100	0.0	NS
San Joaquin River, West of Oulton Point (812)	100	0.0	100	0.0	NS
Sacramento R. across from Sherman Lake (704)	100	0.0	100	0.0	NS

	MSD	PMSD
One-way ANOVA	4.2	4.2
Two-way ANOVA	3.9	3.9

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO ²
DIEPAMHR	0.139	0.012	0.112	0.002	NS
Suisun Bay, East of middle point (504)	0.182	0.011	0.139	0.012	NS
Middle of Broad Slough, West end (804)	0.158	0.017	0.146	0.014	NS
San Joaquin River, West of Oulton Point (812)	0.155	0.005	0.152	0.023	NS
Sacramento R. across from Sherman Lake (704)	0.153	0.012	0.147	0.031	NS

	MSD	PMSD
One-way ANOVA	0.057	40.9
Two-way ANOVA	0.056	40.4

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

Table A36-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 08/23/2006 - 08/24/2006.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Suisun Bay, East of middle point (504)	5630	20.2	7.65	8.72	25.8	0.11	0.0015
Middle of Broad Slough, West end (804)	312	20.5	7.59	8.04	10.4	0.09	0.0013
San Joaquin River, West of Oulton Point (812)	314	21.0	7.59	8.15	10.6	0.05	0.0008
Sacramento R. across from Sherman Lake (704)	598	20.5	7.63	8.46	31.5	0.16	0.0025

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Table A36-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 08/25/2006 of samples collected by the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 08/23/2006 - 08/24/2006.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	361	23.9	23.9	6.4	8.6	7.51	8.02	106	59	-
Suisun Bay, East of middle point (504)	5305	23.8	23.8	6.3	8.5	7.38	7.76	550	70	0.002
Middle of Broad Slough, West end (804)	595	23.8	23.9	6.1	8.5	7.50	7.97	92	60	0.004
San Joaquin River, West of Oulton Point (812)	321	23.8	24.0	5.8	8.5	7.47	7.95	16	62	0.002
Sacramento R. across from Sherman Lake (704)	305	24.0	24.3	6.2	8.6	7.48	7.91	72	62	0.006
DIEPAMHR + 25 ppb BPO	354	23.7	23.8	6.7	8.6	7.51	8.01	-	-	-
Suisun Bay, East of middle point (504) + 25 ppb PBO	5250	23.8	23.9	6.5	8.6	7.37	7.76	-	-	-
Middle of Broad Slough, West end (804) + 25 ppb PBO	579	23.8	23.9	6.2	8.6	7.54	7.98	-	-	-
San Joaquin River, West of Oulton Point (812) + 25 ppb PBO	320	23.9	24.0	6.3	8.6	7.53	7.98	-	-	-
Sacramento R. across from Sherman Lake (704) + 25 ppb PBO	310	23.9	24.1	6.1	8.6	7.50	8.00	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A37-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 9/07/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 9/05/06 - 9/06/06.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	100	0.0	97	3.0	NS
High EC Control @ 16.33 mS/cm	93	4.4	86	3.2	NS
San Joaquin River, West of Oulton Point (812)	96	2.3	100	0.0	NS
Sacramento R. across from Sherman Lake (704)	98	2.0	100	0.0	NS
Old River, western arm at railroad bridge (902)	98	2.0	100	0.0	NS
San Joaquin River between Hog and Turner Cuts (910)	100	0.0	100	0.0	NS
Old River at mouth of Holland Cut (915)	100	0.0	100	0.0	NS
Sacramento Deep Water Channel, Light 55	100	0.0	100	0.0	NS
Sacramento River at tip of Grand Island (711)	96	2.3	100	0.0	NS
Carquinez Strait, West of Benicia army dock (405) ³	94	2.4	85	9.7	NS
Field Dup.: Sacramento Deep Water Channel, Light 55	100	0.0	100	0.0	NS

	MSD	PMSD
One-way ANOVA	7.8	7.8
Two-way ANOVA	13.2	13.2

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.089	0.007	0.058	0.004	NS
High EC Control @ 16.33 mS/cm	0.058	0.004	0.040	0.007	NS
San Joaquin River, West of Oulton Point (812)	0.095	0.007	0.096	0.018	NS
Sacramento R. across from Sherman Lake (704)	0.105	0.010	0.092	0.005	NS
Old River, western arm at railroad bridge (902)	0.131	0.011	0.088	0.012	NS
San Joaquin River between Hog and Turner Cuts (910)	0.124	0.014	0.098	0.018	NS
Old River at mouth of Holland Cut (915)	0.139	0.014	0.088	0.006	NS
Sacramento Deep Water Channel, Light 55	0.132	0.010	0.112	0.012	NS
Sacramento River at tip of Grand Island (711)	0.090	0.005	0.071	0.009	NS
Carquinez Strait, West of Benicia army dock (405) ³	0.082	0.004	0.056	0.010	NS
Field Dup.: Sacramento Deep Water Channel, Light 55	0.131	0.007	0.099	0.011	NS

	MSD	PMSD
One-way ANOVA	0.053	60.2
Two-way ANOVA	0.053	60.2

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. This high conductivity sample was compared to the high EC control.

Table A37-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 9/05/06 - 9/06/06.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
San Joaquin River, West of Oulton Point (812)	209	21.1	7.49	7.3	7.1	0.12	0.001
Sacramento R. across from Sherman Lake (704)	5010	21.4	7.54	7.4	11.0	0.17	0.002
Old River, western arm at railroad bridge (902)	293	21.1	8.31	8.0	3.6	0.08	0.006
San Joaquin River between Hog and Turner Cuts (910)	417	21.6	7.17	6.0	11.5	0.00	0.000
Old River at mouth of Holland Cut (915)	264	21.6	7.99	7.1	4.7	0.05	0.002
Sacramento Deep Water Channel, Light 55	338	21.9	7.85	7.3	29.9	0.00	0.000
Sacramento River at tip of Grand Island (711)	164	21.5	7.41	7.4	6.1	0.15	0.002
Carquinez Strait, West of Benicia army dock (405)	15260	18.9	7.66	7.5	60.7	0.16	0.002
Field Dup.: Sacramento Deep Water Channel, Light 55	338	21.9	7.85	7.3	29.9	0.05	0.001

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Table A37-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 9/07/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 9/05/06 - 9/06/06.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	362	21.9	24.4	6.7	8.3	7.63	8.10	110	59	-
High EC Control @ 16.33 mS/cm	15965	22.0	24.4	6.6	8.5	7.49	7.98	1900	160	-
San Joaquin River, West of Oulton Point (812)	374	22.0	24.6	6.1	8.5	7.69	7.99	62	64	0.006
Sacramento R. across from Sherman Lake (704)	223	22.0	24.7	6.3	8.5	7.65	8.00	68	67	0.008
Old River, western arm at railroad bridge (902)	285	21.9	24.5	6.3	8.6	7.60	8.51	68	61	0.011
San Joaquin River between Hog and Turner Cuts (910)	384	21.8	24.6	6.4	8.8	7.60	7.81	110	71	0.000
Old River at mouth of Holland Cut (915)	288	21.8	24.5	6.5	8.5	7.61	8.15	72	61	0.003
Sacramento Deep Water Channel, Light 55	339	22.0	24.6	6.5	8.3	7.75	8.11	104	91	0.000
Sacramento River at tip of Grand Island (711)	166	21.9	24.6	6.1	8.4	7.65	7.97	70	68	0.007
Carquinez Strait, West of Benicia army dock (405)	14775	21.7	24.6	6.7	8.2	7.51	7.85	1640	128	0.004
Field Dup.: Sacramento Deep Water Channel, Light 55	343	22.0	24.6	6.3	8.6	7.72	8.14	100	88	0.003
DIEPAMHR + 25 ppb PBO	324	21.9	24.2	6.9	8.4	7.73	8.26	-	-	-
High EC Control @ 16.33 mS/cm + 25 ppb PBO	15635	22.3	23.8	6.6	8.8	7.49	7.99	-	-	-
San Joaquin River, West of Oulton Point (812) + 25 ppb PBO	252	22.1	23.9	5.9	8.6	7.63	7.87	-	-	-
Sacramento R. across from Sherman Lake (704) + 25 ppb PBO	197	22.1	24.0	6.2	8.7	7.66	7.90	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	285	22.2	23.8	6.2	8.9	7.60	8.51	-	-	-
San Joaquin River between Hog and Turner Cuts (910) + 25 ppb PBO	392	22.2	23.7	6.3	8.5	7.58	7.78	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	271	22.2	23.7	6.2	8.4	7.59	8.17	-	-	-
Sacramento Deep Water Channel, Light 55 + 25 ppb PBO	341	22.3	23.8	6.4	8.4	7.76	8.14	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	173	22.4	23.8	6.3	8.9	7.63	8.14	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	14815	22.6	23.7	6.8	8.5	7.51	7.78	-	-	-
Field Dup.: Sacramento Deep Water Channel, Light 55 + 25 ppb PBO	342.9	22.5	23.8	6.4	8.5	7.77	8.25	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A38-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 9/08/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 9/06/06 - 9/07/06.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	100	0.0	100	0.0	NS
High EC Control @ 13.01 mS/cm	96	2.3	90	10.0	NS
High EC Control @ 19.82 mS/cm	92	4.9	67	28.5	NS
Suisun Bay, East of middle point (504)	100	0.0	100	0.0	NS
Middle of Broad Slough, West end (804)	98	2.0	100	0.0	NS
Grizzly Bay at Dolphin (602) ³	100	0.0	96	3.7	NS
Suisun Bay off Chipps Island (508)	100	0.0	100	0.0	NS
Montezuma Slough at Nurse Slough (609)	100	0.0	100	0.0	NS
Napa River at Riverside Blvd. terminus (340) ⁴	91	4.6	89	6.4	NS
Bottle Blank	96	2.4	-	-	NA

	MSD	PMSD
One-way ANOVA	12.9	12.9
Two-way ANOVA	29.5	29.5

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.093	0.010	0.054	0.012	NS
High EC Control @ 13.01 mS/cm	0.054	0.007	0.040	0.005	NS
High EC Control @ 19.82 mS/cm	0.035	0.006	0.029	0.005	NS
Suisun Bay, East of middle point (504)	0.092	0.009	0.124	0.006	NS
Middle of Broad Slough, West end (804)	0.106	0.005	0.120	0.006	NS
Grizzly Bay at Dolphin (602) ³	0.081	0.005	0.127	0.001	NS
Suisun Bay off Chipps Island (508)	0.093	0.009	0.115	0.016	NS
Montezuma Slough at Nurse Slough (609)	0.093	0.020	0.125	0.017	NS
Napa River at Riverside Blvd. terminus (340) ⁴	0.020	0.005	0.060	0.014	NS
Bottle Blank	0.043	0.008	-	-	NA

	MSD	PMSD
One-way ANOVA	0.054	58.0
Two-way ANOVA	0.053	56.6

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. This high conductivity sample was compared to the High EC Control @ 13.01 mS/cm.

4. This high conductivity sample was compared to the High EC Control @ 19.82 mS/cm.

Table A38-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 9/06/06 - 9/07/06.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Suisun Bay, East of middle point (504)	4346	19.3	7.68	7.6	19.6	0.15	0.002
Middle of Broad Slough, West end (804)	518	20.8	8.00	7.5	8.3	0.10	0.004
Grizzly Bay at Dolphin (602)	12780	18.5	7.64	7.7	122.0	0.18	0.002
Suisun Bay off Chipps Island (508)	1850	19.8	7.60	7.6	14.0	0.13	0.002
Montezuma Slough at Nurse Slough (609)	5001	20.2	7.49	7.4	41.4	0.08	0.001
Napa River at Riverside Blvd. terminus (340)	19140	21.7	7.42	6.3	11.8	0.05	0.000
Bottle Blank	-	-	-	-	-	0.00	-

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Table A38-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 9/08/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 9/06/06 - 9/07/06.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	358	22.3	24.0	7.1	8.2	7.53	8.13	110	59	-
High EC Control @ 13.01 mS/cm	12575	22.2	23.8	6.7	8.2	7.55	8.00	1500	140	-
High EC Control @ 19.82 mS/cm	19375	22.3	24.1	7.3	8.5	7.62	8.04	3080	160	-
Suisun Bay, East of middle point (504)	4955	22.4	23.8	6.9	8.7	7.59	8.04	532	74	0.006
Middle of Broad Slough, West end (804)	564	22.3	23.7	6.7	8.5	7.65	8.19	96	70	0.007
Grizzly Bay at Dolphin (602)	12020	22.3	24.0	6.9	8.4	7.55	7.96	1400	140	0.006
Suisun Bay off Chipps Island (508)	1672	22.4	23.9	6.6	8.4	7.61	8.12	210	66	0.007
Montezuma Slough at Nurse Slough (609)	4850	22.2	23.9	7.2	8.3	7.62	8.06	556	82	0.004
Napa River at Riverside Blvd. terminus (340)	18410	22.3	23.7	7.2	8.6	7.86	7.93	3160	280	0.001
Bottle Blank	469	22.4	23.9	7.2	8.6	7.78	8.18	114	59	0.000
DIEPAMHR + 25 ppb PBO	368	22.4	24.0	6.9	8.1	7.70	8.15	-	-	-
High EC Control @ 13.01 mS/cm + 25 ppb PBO	12695	22.4	23.9	7.1	8.5	7.51	8.00	-	-	-
High EC Control @ 19.82 mS/cm + 25 ppb PBO	19080	22.5	23.7	7.4	8.6	7.61	8.05	-	-	-
Suisun Bay, East of middle point (504) + 25 ppb PBO	4800	22.5	23.7	7.2	8.4	7.64	8.08	-	-	-
Middle of Broad Slough, West end (804) + 25 ppb PBO	576	22.5	23.6	6.9	8.6	7.72	8.17	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	12120	22.5	23.8	7.2	8.6	7.58	7.98	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	9235	22.5	23.7	6.8	8.6	7.70	8.13	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	4837	22.4	23.5	7.3	8.4	7.67	8.01	-	-	-
Napa River at Riverside Blvd. terminus (340) + 25 ppb PBO	18280	22.5	23.6	7.3	8.5	7.86	7.98	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A39-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 9/21/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 9/19/06 - 9/20/06.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	98	1.8	100	0.0	NS
High EC Control @ 22.13 mS/cm	87	4.0	68	1.1	NS
Napa River at Riverside Blvd. terminus (340) ³	84	11.7	87	8.8	NS
Old River, western arm at railroad bridge (902)	100	0.0	100	0.0	NS
Old River at mouth of Holland Cut (915)	100	0.0	100	0.0	NS
San Joaquin River between Hog and Turner Cuts (910)	100	0.0	97	3.0	NS
San Joaquin River, West of Oulton Point (812)	100	0.0	100	0.0	NS
Sacramento River at tip of Grand Island (711)	98	1.8	100	0.0	NS
Sacramento Deep Water Channel, Light 55	98	1.8	100	0.0	NS
Sacramento R. across from Sherman Lake (704)	98	2.0	97	3.3	NS
Middle of Broad Slough, West end (804)	96	2.4	100	0.0	NS
Field Dup.: San Joaquin River, West of Oulton Point (812)	100	0.0	97	3.3	NS

	MSD	PMSD
One-way ANOVA	21.2	21.6
Two-way ANOVA	21.2	21.6

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.096	0.005	0.084	0.005	NS
High EC Control @ 22.13 mS/cm	0.073	0.003	0.041	0.004	NS
Napa River at Riverside Blvd. terminus (340) ³	0.086	0.006	0.051	0.011	NS
Old River, western arm at railroad bridge (902)	0.154	0.007	0.124	0.003	NS
Old River at mouth of Holland Cut (915)	0.146	0.011	0.113	0.014	NS
San Joaquin River between Hog and Turner Cuts (910)	0.149	0.008	0.130	0.008	NS
San Joaquin River, West of Oulton Point (812)	0.135	0.008	0.109	0.005	NS
Sacramento River at tip of Grand Island (711)	0.113	0.008	0.102	0.006	NS
Sacramento Deep Water Channel, Light 55	0.125	0.009	0.127	0.007	NS
Sacramento R. across from Sherman Lake (704)	0.120	0.007	0.107	0.003	NS
Middle of Broad Slough, West end (804)	0.144	0.006	0.125	0.006	NS
Field Dup.: San Joaquin River, West of Oulton Point (812)	0.140	0.009	0.134	0.005	NS

	MSD	PMSD
One-way ANOVA	0.041	43.1
Two-way ANOVA	0.041	43.1

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. This high conductivity sample was compared to the High EC Control @ 22.13 mS/cm.

Table A39-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 9/19/06 - 9/20/06.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Napa River at Riverside Blvd. terminus (340)	21580	19.8	7.72	6.4	9.9	0.07	0.001
Old River, western arm at railroad bridge (902)	311	20.4	8.72	8.7	3.9	0.04	0.007
Old River at mouth of Holland Cut (915)	304	20.6	8.17	8.0	5.2	0.05	0.003
San Joaquin River between Hog and Turner Cuts (910)	567	20.6	7.47	6.0	13.0	0.23	0.003
San Joaquin River, West of Oulton Point (812)	210	19.9	7.83	7.8	6.5	0.07	0.002
Sacramento River at tip of Grand Island (711)	200	18.1	7.81	8.3	13.1	0.19	0.004
Sacramento Deep Water Channel, Light 55	335	19.6	8.02	8.2	37.6	0.06	0.002
Sacramento R. across from Sherman Lake (704)	182	19.5	7.83	7.9	10.6	0.11	0.003
Middle of Broad Slough, West end (804)	604	20.4	8.10	8.2	10.6	0.06	0.003
Field Dup.: San Joaquin River, West of Oulton Point (812)	210	19.9	7.83	7.8	33.6	0.05	0.001

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Table A39-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 9/21/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 9/19/06 - 9/20/06.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	340	21.8	24.6	6.5	8.3	7.51	8.11	108	58	-
High EC Control @ 22.13 mS/cm	21150	22.0	24.6	7.1	8.9	7.56	7.79	2720	180	-
Napa River at Riverside Blvd. terminus (340)	20470	22.1	24.7	6.5	8.7	7.64	7.86	2720	220	0.001
Old River, western arm at railroad bridge (902)	280	21.9	24.6	6.4	8.6	7.82	8.49	68	68	0.005
Old River at mouth of Holland Cut (915)	264	22.0	24.9	6.5	8.9	7.77	8.16	66	67	0.003
San Joaquin River between Hog and Turner Cuts (910)	395	21.7	24.8	6.2	8.6	7.68	8.00	96	76	0.006
San Joaquin River, West of Oulton Point (812)	215	21.8	24.7	6.2	8.6	7.71	8.01	68	72	0.003
Sacramento River at tip of Grand Island (711)	182	21.8	24.7	6.6	8.7	7.70	7.99	62	72	0.009
Sacramento Deep Water Channel, Light 55	328	21.8	24.7	6.5	8.5	7.72	8.03	96	90	0.003
Sacramento R. across from Sherman Lake (704)	194	21.9	24.6	6.6	8.7	7.65	8.01	48	74	0.006
Middle of Broad Slough, West end (804)	567	21.7	24.6	6.4	8.7	7.58	8.00	118	67	0.003
Field Dup.: San Joaquin River, West of Oulton Point (812)	339	21.7	24.6	6.7	8.6	7.70	8.11	102	89	0.003
DIEPAMHR + 25 ppb PBO	345	21.8	24.4	6.8	8.2	7.75	8.14	-	-	-
High EC Control @ 22.13 mS/cm + 25 ppb PBO	20980	21.7	24.3	7.3	8.6	7.43	7.87	-	-	-
Napa River at Riverside Blvd. terminus (340) + 25 ppb PBO	20360	21.5	24.1	6.6	8.6	7.70	7.79	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	281	21.4	24.1	6.8	8.8	8.01	8.56	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	262	21.6	24.1	6.7	8.6	7.88	8.15	-	-	-
San Joaquin River between Hog and Turner Cuts (910) + 25 ppb PBO	406	21.3	24.0	6.8	8.5	7.86	7.97	-	-	-
San Joaquin River, West of Oulton Point (812) + 25 ppb PBO	217	21.6	24.0	6.5	8.7	7.77	8.04	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	183	21.2	24.3	6.7	8.7	7.76	8.02	-	-	-
Sacramento Deep Water Channel, Light 55 + 25 ppb PBO	328.8	21.4	24.0	6.2	8.4	7.77	8.13	-	-	-
Sacramento R. across from Sherman Lake (704) + 25 ppb PBO	196	21.5	24.0	6.5	8.6	7.72	8.02	-	-	-
Middle of Broad Slough, West end (804) + 25 ppb PBO	577	21.6	24.0	6.3	8.7	7.61	8.01	-	-	-
Field Dup.: San Joaquin River, West of Oulton Point (812) + 25 ppb PBO	340	21.8	24.0	6.7	8.7	7.77	8.17	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A40-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 9/22/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 9/21/06.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	100	0.0	97	3.3	NS
Suisun Bay off Chipps Island (508)	94	2.5	97	3.3	NS
Suisun Bay, East of middle point (504)	96	4.0	97	3.0	NS
Carquinez Strait, West of Benicia army dock (405)	96	2.6	97	3.3	NS
Grizzly Bay at Dolphin (602)	100	0.0	97	3.3	NS
Montezuma Slough at Nurse Slough (609)	100	0.0	97	3.3	NS
Bottle Blank	96	2.3	-	-	NA

	MSD	PMSD
One-way ANOVA	11.2	11.2
Two-way ANOVA	13.6	13.6

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.064	0.006	0.045	0.004	NS
Suisun Bay off Chipps Island (508)	0.119	0.005	0.065	0.010	S (55%)
Suisun Bay, East of middle point (504)	0.115	0.011	0.054	0.009	S (47%)
Carquinez Strait, West of Benicia army dock (405)	0.101	0.007	0.054	0.009	S (53%)
Grizzly Bay at Dolphin (602)	0.085	0.004	0.067	0.009	NS
Montezuma Slough at Nurse Slough (609)	0.129	0.008	0.095	0.016	NS
Bottle Blank	0.095	0.006	-	-	NA

	MSD	PMSD
One-way ANOVA	0.034	52.6
Two-way ANOVA	0.040	62.1

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

Table A40-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 09/21/06.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Suisun Bay off Chipps Island (508)	2725	19.8	7.87	8.0	19.3	0.18	0.004
Suisun Bay, East of middle point (504)	6200	19.5	7.92	7.8	28.8	0.10	0.002
Carquinez Strait, West of Benicia army dock (405)	14500	19.1	7.91	7.8	58.3	0.07	0.001
Grizzly Bay at Dolphin (602)	9160	19.2	8.03	8.2	12.8	0.05	0.001
Montezuma Slough at Nurse Slough (609)	6930	20.0	7.87	7.8	61.0	0.04	0.001
Bottle Blank	-	-	-	-	0.5	0.00	-

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Table A40-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 9/22/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 9/21/06.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	351	22.4	23.2	6.6	8.3	7.64	8.16	108	58	-
Suisun Bay off Chipps Island (508)	2478	22.5	23.6	6.8	8.3	7.60	7.85	296	68	0.005
Suisun Bay, East of middle point (504)	5630	22.6	23.5	6.5	8.2	7.59	7.88	658	70	0.003
Carquinez Strait, West of Benicia army dock (405)	13520	22.6	23.0	6.7	8.2	7.54	7.77	1560	78	0.001
Grizzly Bay at Dolphin (602)	8615	22.7	23.7	6.6	8.3	7.60	7.91	980	73	0.001
Montezuma Slough at Nurse Slough (609)	6455	22.6	23.2	6.9	8.2	7.75	7.83	750	80	0.001
Bottle Blank	435	22.6	23.1	6.8	8.1	7.79	8.25	110	58	0.000
DIEPAMHR + 25 ppb PBO	358	22.5	22.6	7.0	8.1	7.75	8.17	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	2550	22.6	22.9	7.1	8.0	7.59	8.03	-	-	-
Suisun Bay, East of middle point (504) + 25 ppb PBO	5720	22.6	22.7	6.5	8.0	7.54	7.95	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	13190	22.6	22.7	6.7	8.0	7.53	7.76	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	8335	22.5	22.7	7.0	8.0	7.66	7.92	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	6305	22.6	22.7	6.8	8.0	7.76	7.95	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A41-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 10/05/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 10/03/06 - 10/04/06.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO ²
DIEPAMHR	96	2.4	100	0.0	NS
Old River, western arm at railroad bridge (902)	94	2.4	93	6.7	NS
Old River at mouth of Holland Cut (915)	100	0.0	100	0.0	NS
San Joaquin River between Hog and Turner Cuts (910)	100	0.0	100	0.0	NS
San Joaquin River, West of Oulton Point (812)	94	2.5	97	3.3	NS
Sacramento River at tip of Grand Island (711)	98	2.0	90	5.8	NS
Sacramento Deep Water Channel, Light 55	98	2.0	100	0.0	NS
Sacramento R. across from Sherman Lake (704)	98	2.0	100	0.0	NS
Montezuma Slough at Nurse Slough (609)	100	0.0	97	3.3	NS
Grizzly Bay at Dolphin (602)	98	2.0	100	0.0	NS

	MSD	PMSD
One-way ANOVA	9.8	10.2
Two-way ANOVA	12.2	12.7

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO ²
DIEPAMHR	0.072	0.001	0.065	0.004	NS
Old River, western arm at railroad bridge (902)	0.103	0.005	0.072	0.000	S (71%)
Old River at mouth of Holland Cut (915)	0.101	0.006	0.079	0.004	NS
San Joaquin River between Hog and Turner Cuts (910)	0.109	0.005	0.078	0.003	S (72%)
San Joaquin River, West of Oulton Point (812)	0.083	0.004	0.082	0.005	NS
Sacramento River at tip of Grand Island (711)	0.069	0.002	0.041	0.004	S (55%)
Sacramento Deep Water Channel, Light 55	0.093	0.004	0.077	0.006	NS
Sacramento R. across from Sherman Lake (704)	0.076	0.004	0.096	0.011	NS
Montezuma Slough at Nurse Slough (609)	0.078	0.007	0.105	0.005	NS
Grizzly Bay at Dolphin (602)	0.073	0.003	0.082	0.004	NS

	MSD	PMSD
One-way ANOVA	0.024	33.2
Two-way ANOVA	0.026	35.7

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

Table A41-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 10/03/06 - 10/04/06.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Old River, western arm at railroad bridge (902)	275	18.0	8.2	7.3	2.9	0.20	0.010
Old River at mouth of Holland Cut (915)	244	18.1	7.9	6.8	2.8	0.22	0.006
San Joaquin River between Hog and Turner Cuts (910)	372	18.7	7.5	5.5	9.1	0.26	0.003
San Joaquin River, West of Oulton Point (812)	170	18.0	7.76	6.7	3.9	0.29	0.005
Sacramento River at tip of Grand Island (711)	133	18.0	7.68	6.8	3.2	0.08	0.001
Sacramento Deep Water Channel, Light 55	295	18.2	7.68	6.6	25.5	0.24	0.004
Sacramento R. across from Sherman Lake (704)	278	17.5	7.75	6.8	19.5	0.30	0.005
Montezuma Slough at Nurse Slough (609)	6380	17.0	7.68	7.7	34.9	0.23	0.003
Grizzly Bay at Dolphin (602)	8980	17.0	7.86	7.5	39.9	0.27	0.005

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Table A41-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 10/05/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 10/03/06 - 10/04/06.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	314	22.0	23.3	6.5	8.4	7.64	8.05	108	59	-
Old River, western arm at railroad bridge (902)	231	22.1	23.2	6.4	8.6	7.65	8.06	76	67	0.010
Old River at mouth of Holland Cut (915)	212	22.2	22.9	6.4	8.6	7.35	7.95	72	68	0.009
San Joaquin River between Hog and Turner Cuts (910)	340	22.1	23.7	6.5	8.8	7.61	7.86	96	70	0.009
San Joaquin River, West of Oulton Point (812)	158	22.1	23.5	6.5	8.5	7.64	7.93	60	60	0.011
Sacramento River at tip of Grand Island (711)	123	22.0	23.5	6.4	8.7	7.58	7.87	56	58	0.003
Sacramento Deep Water Channel, Light 55	262	22.0	23.5	6.4	8.9	7.71	8.04	96	84	0.012
Sacramento R. across from Sherman Lake (704)	243	22.0	23.4	6.4	8.6	7.68	7.91	80	66	0.011
Montezuma Slough at Nurse Slough (609)	8040	22.0	23.6	6.8	8.9	7.62	7.78	188	86	0.005
Grizzly Bay at Dolphin (602)	5690	22.0	23.6	6.7	8.5	7.54	7.77	268	76	0.006
DIEPAMHR + 25 ppb PBO	306	22.0	23.2	7.1	8.5	7.74	8.05	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	222	21.9	23.8	6.5	8.8	7.41	8.09	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	218	21.9	23.9	7.2	8.8	7.48	8.05	-	-	-
San Joaquin River between Hog and Turner Cuts (910) + 25 ppb PBO	336	21.8	23.9	6.7	8.9	7.42	7.96	-	-	-
San Joaquin River, West of Oulton Point (812) + 25 ppb PBO	160	21.7	23.9	6.9	8.6	7.47	8.05	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	125	21.8	23.6	7.2	8.6	7.49	7.88	-	-	-
Sacramento Deep Water Channel, Light 55 + 25 ppb PBO	261	21.7	24.0	7.3	8.7	7.71	8.04	-	-	-
Sacramento R. across from Sherman Lake (704) + 25 ppb PBO	245	21.5	24.1	7.2	8.6	7.56	8.01	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	5670	21.6	24.0	7.2	8.7	7.49	7.83	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	8180	21.5	23.9	7.3	8.8	7.10	7.80	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A42-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 10/6/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 10/4/06 - 10/5/06.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	84	15.6	100	0.0	NS
High EC Control @ 18.16 mS/cm	88	3.5	68	5.2	NS
High EC Control @ 21.25 mS/cm	64	7.2	57	28.5	NS
Carquinez Strait, West of Benicia army dock (405) ³	85	7.6	89	6.4	NS
Suisun Bay, East of middle point (504)	96	4.0	100	0.0	NS
Suisun Bay off Chipps Island (508)	100	0.0	96	3.7	NS
Middle of Broad Slough, West end (804)	100	0.0	96	3.7	NS
Napa River at Riverside Blvd. terminus (340) ⁴	75	4.9	41	23.1	NS
Field Dup.: Napa River at Riverside Blvd. terminus (340) ⁴	92	3.5	-	-	NA

	MSD	PMSD
One-way ANOVA	35.6	42.1
Two-way ANOVA	36.5	43.2

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.065	0.010	0.054	0.003	NS
High EC Control @ 18.16 mS/cm	0.034	0.003	0.044	0.006	NS
High EC Control @ 21.25 mS/cm	0.031	0.007	0.035	0.003	NS
Carquinez Strait, West of Benicia army dock (405) ³	0.046	0.004	0.050	0.003	NS
Suisun Bay, East of middle point (504)	0.077	0.008	0.057	0.006	NS
Suisun Bay off Chipps Island (508)	0.085	0.002	0.075	0.001	NS
Middle of Broad Slough, West end (804)	0.091	0.006	0.078	0.016	NS
Napa River at Riverside Blvd. terminus (340) ⁴	0.046	0.004	0.052	0.011	NS
Field Dup.: Napa River at Riverside Blvd. terminus (340) ⁴	0.042	0.005	-	-	NA

	MSD	PMSD
One-way ANOVA	0.031	47.3
Two-way ANOVA	0.034	51.8

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. This high conductivity sample was compared to the High EC control at 18.16 mS/cm.

4. This high conductivity sample was compared to the High EC control at 21.25 mS/cm.

Table A42-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 10/4/06 - 10/05/06.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Carquinez Strait, West of Benicia army dock (405)	18010	17.1	7.8	7.3	23.9	0.23	0.003
Suisun Bay, East of middle point (504)	4602	17.5	7.88	7.6	13.5	0.00	0.000
Suisun Bay off Chipps Island (508)	1738	18.4	7.91	7.4	15.2	0.24	0.006
Middle of Broad Slough, West end (804)	488	18.2	8.09	7.3	7.6	0.22	0.008
Napa River at Riverside Blvd. terminus (340)	21400	17.8	7.61	5.8	7.5	0.02	0.000
Field Dup.: Napa River at Riverside Blvd. terminus (340)	21400	17.8	7.61	5.8	8.2	0.03	0.000

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Table A42-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 10/6/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 10/4/06 - 10/5/06.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	325	22.3	23.6	7.3	8.3	7.76	8.09	112	59	-
High EC Control @ 18.16 mS/cm	17405	22.2	23.5	7.7	8.7	7.62	7.89	2300	44	-
High EC Control @ 21.25 mS/cm	19735	22.3	23.5	7.8	8.4	7.63	7.95	2700	84	-
Carquinez Strait, West of Benicia army dock (405)	16410	22.3	23.8	7.1	8.8	7.62	7.82	2128	88	0.005
Suisun Bay, East of middle point (504)	4284	22.2	23.5	7.2	8.8	7.60	7.92	592	76	0.000
Suisun Bay off Chipps Island (508)	1733	22.1	23.8	7.3	8.6	7.67	8.05	256	70	0.011
Middle of Broad Slough, West end (804)	478	22.2	23.5	7.0	8.7	7.80	8.18	104	71	0.014
Napa River at Riverside Blvd. terminus (340)	19730	22.2	23.6	7.3	8.9	7.55	7.96	2790	142	0.000
Field Dup.: Napa River at Riverside Blvd. terminus (340)	19555	22.0	23.4	7.5	8.8	7.64	8.00	2780	140	0.000
DIEPAMHR + 25 ppb PBO	323	22.2	23.7	7.2	8.3	7.76	8.17	-	-	-
High EC Control @ 18.16 mS/cm + 25 ppb PBO	17325	22.3	23.7	7.4	8.5	7.58	7.91	-	-	-
High EC Control @ 21.25 mS/cm + 25 ppb PBO	20225	22.3	23.6	7.6	8.6	7.61	7.93	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	16440	22.3	23.7	7.4	8.4	7.62	7.82	-	-	-
Suisun Bay, East of middle point (504) + 25 ppb PBO	4395	22.1	23.6	7.2	8.9	7.78	8.03	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	1717	22.2	23.6	7.3	8.3	7.87	8.06	-	-	-
Middle of Broad Slough, West end (804) + 25 ppb PBO	524	22.2	23.6	7.2	8.6	7.92	8.14	-	-	-
Napa River at Riverside Blvd. terminus (340) + 25 ppb PBO	18805	22.1	23.7	7.4	8.5	7.57	7.95	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A43-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 10/19/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 10/17/06 - 10/18/06.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	100	0.0	100	0.0	NS
Sacramento R. across from Sherman Lake (704)	100	0.0	100	0.0	NS
San Joaquin River, West of Oulton Point (812)	96	3.6	100	0.0	NS
Old River at mouth of Holland Cut (915)	100	0.0	100	0.0	NS
Sacramento River at tip of Grand Island (711)	96	2.4	93	3.3	NS
Old River, western arm at railroad bridge (902)	100	0.0	100	0.0	NS
San Joaquin River between Hog and Turner Cuts (910)	100	0.0	100	0.0	NS
Middle of Broad Slough, West end (804)	100	0.0	100	0.0	NS
Sacramento Deep Water Channel, Light 55	100	0.0	100	0.0	NS
Suisun Bay off Chipps Island (508)	98	2.0	100	0.0	NS

	MSD	PMSD
One-way ANOVA	9.0	9.0
Two-way ANOVA	8.6	8.6

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.059	0.005	0.062	0.005	NS
Sacramento R. across from Sherman Lake (704)	0.096	0.004	0.099	0.010	NS
San Joaquin River, West of Oulton Point (812)	0.075	0.005	0.088	0.004	NS
Old River at mouth of Holland Cut (915)	0.074	0.004	0.084	0.004	NS
Sacramento River at tip of Grand Island (711)	0.063	0.005	0.067	0.005	NS
Old River, western arm at railroad bridge (902)	0.085	0.006	0.087	0.008	NS
San Joaquin River between Hog and Turner Cuts (910)	0.090	0.005	0.102	0.003	NS
Middle of Broad Slough, West end (804)	0.093	0.004	0.095	0.004	NS
Sacramento Deep Water Channel, Light 55	0.091	0.006	0.080	0.008	NS
Suisun Bay off Chipps Island (508)	0.086	0.007	0.071	0.002	NS

	MSD	PMSD
One-way ANOVA	0.028	47.3
Two-way ANOVA	0.030	50.7

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

Table A43-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 10/17/06 - 10/18/06.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Sacramento R. across from Sherman Lake (704)	813	17.8	7.28	8.9	19.5	0.05	0.000
San Joaquin River, West of Oulton Point (812)	191	17.0	7.37	9.1	4.2	0.27	0.002
Old River at mouth of Holland Cut (915)	244	17.5	7.38	8.7	2.5	0.24	0.002
Sacramento River at tip of Grand Island (711)	138	16.9	7.31	8.8	3.7	0.54	0.003
Old River, western arm at railroad bridge (902)	288	17.2	7.55	9.1	2.4	0.24	0.003
San Joaquin River between Hog and Turner Cuts (910)	369	17.3	7.15	7.6	9.9	0.35	0.001
Middle of Broad Slough, West end (804)	2226	17.4	7.57	8.9	15.1	0.29	0.003
Sacramento Deep Water Channel, Light 55	268	17.8	7.36	8.7	32.0	0.29	0.002
Suisun Bay off Chipps Island (508)	3006	16.4	7.93	8.4	12.7	0.05	0.001

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Table A43-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 10/19/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 10/17/06 - 10/18/06.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	346	23.0	23.8	7.0	8.5	7.82	8.20	108	61	-
Sacramento R. across from Sherman Lake (704)	680	23.2	23.7	6.7	8.5	7.75	8.10	124	96	0.003
San Joaquin River, West of Oulton Point (812)	204	23.2	24.0	6.5	8.9	7.58	7.92	64	46	0.009
Old River at mouth of Holland Cut (915)	250	23.2	23.9	6.5	8.6	7.64	7.92	80	71	0.009
Sacramento River at tip of Grand Island (711)	150	23.2	23.9	6.2	8.9	7.57	7.86	56	62	0.014
Old River, western arm at railroad bridge (902)	256	23.2	23.8	6.4	8.8	7.56	8.27	74	73	0.020
San Joaquin River between Hog and Turner Cuts (910)	372	23.2	23.7	6.7	8.7	7.68	7.80	98	76	0.009
Middle of Broad Slough, West end (804)	2558	23.2	23.7	7.1	8.8	7.58	7.77	1680	88	0.007
Sacramento Deep Water Channel, Light 55	295	23.2	23.6	6.4	8.8	7.74	7.95	96	92	0.011
Suisun Bay off Chipps Island (508)	2818	23.2	23.6	6.9	8.8	7.57	7.76	376	82	0.001
DIEPAMHR + 25 ppb PBO	377	23.3	23.8	7.3	8.8	7.75	8.24	-	-	-
Sacramento R. across from Sherman Lake (704) + 25 ppb PBO	697	23.4	23.9	6.7	8.4	7.66	8.10	-	-	-
San Joaquin River, West of Oulton Point (812) + 25 ppb PBO	211	23.3	24.0	6.7	8.4	7.67	8.13	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	250	23.5	24.0	6.7	8.4	7.63	8.18	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	235	23.3	24.1	6.8	8.5	7.64	8.11	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	261	23.2	24.0	6.6	8.8	7.58	8.13	-	-	-
San Joaquin River between Hog and Turner Cuts (910) + 25 ppb PBO	367	23.2	23.9	7.5	8.3	7.63	7.92	-	-	-
Middle of Broad Slough, West end (804) + 25 ppb PBO	2458	23.1	24.1	6.9	8.7	7.67	7.88	-	-	-
Sacramento Deep Water Channel, Light 55 + 25 ppb PBO	293	23.3	24.1	6.7	8.6	7.78	8.07	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	2662	23.3	24.1	7.1	8.7	7.62	7.88	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A44-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 10/20/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 10/05/06.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	100	0.0	100	0.0	NS
High EC Control @ 21.25 mS/cm	82	9.2	69	7.1	NS
Napa River at Riverside Blvd Terminous (340) ³	78	7.9	79	0.7	NS

	MSD	PMSD
One-way ANOVA	26.4	26.4
Two-way ANOVA	30.7	30.7

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.055	0.009	0.054	0.013	NS
High EC Control @ 21.25 mS/cm	0.030	0.002	0.031	0.008	NS
Napa River at Riverside Blvd Terminous (340) ³	0.028	0.003	0.024	0.005	NS

	MSD	PMSD
One-way ANOVA	0.021	37.6
Two-way ANOVA	0.031	56.1

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. This high conductivity sample was compared to the High EC control @ 21.25 mS/cm.

Table A44-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 10/05/06.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Napa River at Riverside Blvd Terminous (340)	21400	17.8	7.61	5.8	7.5	0.02	0.000

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Table A44-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 10/20/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 10/05/06.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	352	21.8	24.2	7.6	8.6	7.99	8.24	112	59	-
High EC Control @ 21.25 mS/cm	20405	22.0	24.3	7.9	8.8	7.63	7.82	2300	44	-
Napa River at Riverside Blvd Terminus (340)	20410	22.0	24.3	7.1	8.9	7.50	8.01	2790	142	0.000
DIEPAMHR + PBO	445	22.0	24.0	7.8	8.5	7.90	8.45	-	-	-
High EC Control @ 21.25 mS/cm + 25 ppb PBO	20370	21.9	24.0	7.8	8.7	7.67	7.92	-	-	-
Napa River at Riverside Blvd Terminus (340) + 25 ppb PBO	20095	21.9	24.0	7.7	8.9	7.65	7.95	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A45-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 10/20/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 10/18/06 - 10/19/06.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	100	0.0	100	0.0	NS
High EC Control @ 11.42 mS/cm	100	0.0	90	5.8	NS
High EC Control @ 16.25 mS/cm	86	5.2	86	3.0	NS
High EC Control @ 21.00 mS/cm	72	18.3	68	6.5	NS
Grizzly Bay at Dolphin (602) ³	96	2.5	96	3.7	NS
Suisun Bay, East of middle point (504)	98	2.0	100	0.0	NS
Montezuma Slough at Nurse Slough (609)	100	0.0	97	3.3	NS
Carquinez Strait, West of Benicia army dock (405) ⁴	96	2.4	100	0.0	NS
Napa River at Riverside Blvd. terminus (340) ⁵	80	8.3	62	9.0	NS
Field Dup.: Grizzly Bay at Dolphin (602)	98	2.2	-	-	NA

	MSD	PMSD
One-way ANOVA	32.1	32.1
Two-way ANOVA	22.7	22.7

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.055	0.003	0.055	0.011	NS
High EC Control @ 11.42 mS/cm	0.048	0.005	0.035	0.002	NS
High EC Control @ 16.25 mS/cm	0.044	0.005	0.053	0.008	NS
High EC Control @ 21.00 mS/cm	0.029	0.010	0.056	0.015	NS
Grizzly Bay at Dolphin (602) ³	0.067	0.008	0.051	0.020	NS
Suisun Bay, East of middle point (504)	0.060	0.006	0.051	0.008	NS
Montezuma Slough at Nurse Slough (609)	0.075	0.010	0.075	0.007	NS
Carquinez Strait, West of Benicia army dock (405) ⁴	0.049	0.009	0.055	0.013	NS
Napa River at Riverside Blvd. terminus (340) ⁵	0.043	0.006	0.035	0.007	NS
Field Dup.: Grizzly Bay at Dolphin (602)	0.057	0.008	-	-	NA

	MSD	PMSD
One-way ANOVA	0.034	62.5
Two-way ANOVA	0.046	84.5

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.
3. This high conductivity sample was compared to the High EC control @ 11.42 mS/cm.
4. This high conductivity sample was compared to the High EC control @ 16.25 mS/cm.
5. This high conductivity sample was compared to the High EC control @ 21.00 mS/cm.

Table A45-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 10/18/06 - 10/19/06.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Grizzly Bay at Dolphin (602)	11850	16.1	7.94	8.5	16.7	0.27	0.005
Suisun Bay, East of middle point (504)	6590	16.4	7.92	8.7	1.4	0.26	0.005
Montezuma Slough at Nurse Slough (609)	9220	16.1	7.71	8.4	35.1	0.25	0.003
Carquinez Strait, West of Benicia army dock (405)	17290	16.4	7.85	8.0	19.9	0.29	0.004
Napa River at Riverside Blvd. terminus (340)	22080	16.0	7.59	9.4	5.8	0.08	0.001
Field Dup.: Grizzly Bay at Dolphin (602)	11850	16.1	7.94	8.5	16.7	0.27	0.005

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Table A45-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 10/20/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 10/18/06 - 10/19/06.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	364	22.7	23.6	7.6	8.3	7.80	8.29	104	61	-
High EC Control @11.42 mS/cm	11430	22.7	24.0	7.5	8.9	7.59	7.92	1400	110	-
High EC Control @ 16.25 mS/cm	16355	22.8	23.9	7.8	8.4	7.68	7.95	2040	180	-
High EC Control @ 21.00 mS/cm	20945	22.8	24.0	8.1	8.4	7.75	7.96	1560	140	-
Grizzly Bay at Dolphin (602)	11090	22.7	24.0	7.6	8.5	7.76	7.89	1680	140	0.008
Suisun Bay, East of middle point (504)	5800	22.7	24.0	7.5	8.6	7.81	7.94	1840	110	0.009
Montezuma Slough at Nurse Slough (609)	8440	22.6	23.9	7.8	8.5	7.75	7.81	1220	150	0.005
Carquinez Strait, West of Benicia army dock (405)	15635	22.5	24.2	7.6	8.9	7.64	7.77	2280	110	0.005
Napa River at Riverside Blvd. terminus (340)	20445	22.5	24.3	7.6	8.8	7.71	7.90	3180	240	0.001
Field Dup.: Grizzly Bay at Dolphin (602)	11275	22.4	24.3	7.9	8.9	7.78	8.00	1720	160	0.006
DIEPAMR + 25 ppb PBO	444	23.4	23.4	7.6	8.5	7.85	8.10	-	-	-
High EC Control @11.42 mS/cm + 25 ppb PBO	11535	22.6	24.0	7.5	8.5	7.61	7.90	-	-	-
High EC Control @ 16.25 mS/cm + 25 ppb PBO	15970	24.0	24.0	7.8	8.3	7.73	7.95	-	-	-
High EC Control @ 21.00 mS/cm + 25 ppb PBO	20560	24.1	24.1	8.2	8.4	7.78	7.97	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	11420	24.0	24.0	7.6	8.5	7.77	7.78	-	-	-
Suisun Bay, East of middle point (504) + 25 ppb PBO	5450	23.8	23.8	7.7	8.5	7.86	7.87	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	8220	23.9	23.9	7.9	8.8	7.71	7.85	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	15200	24.0	24.0	7.9	8.8	7.70	7.81	-	-	-
Napa River at Riverside Blvd. terminus (340) + 25 ppb PBO	19950	24.0	24.0	7.7	8.6	7.80	7.95	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A46-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 11/02/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 10/31/06.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO ²
DIEPAMHR	98	2.0	97	3.3	NS
Old River, western arm at railroad bridge (902)	100	0.0	100	0.0	NS
Old River at mouth of Holland Cut (915)	95	5.0	100	0.0	NS
San Joaquin River between Hog and Turner Cuts (910)	100	0.0	100	0.0	NS
San Joaquin River, West of Oulton Point (812)	100	0.0	100	0.0	NS
Sacramento Deep Water Channel, Light 55	100	0.0	100	0.0	NS
Sacramento River at tip of Grand Island (711)	100	0.0	100	0.0	NS
Sacramento R. across from Sherman Lake (704)	100	0.0	97	3.3	NS
Middle of Broad Slough, West end (804)	98	2.0	100	0.0	NS
Field Dup.: Sacramento River at tip of Grand Island (711)	96	2.4	-	-	NA

	MSD	PMSD
One-way ANOVA	9.4	3.5
Two-way ANOVA	10.2	10.4

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO ²
DIEPAMHR	0.059	0.003	0.059	0.013	NS
Old River, western arm at railroad bridge (902)	0.082	0.005	0.085	0.006	NS
Old River at mouth of Holland Cut (915)	0.087	0.013	0.082	0.015	NS
San Joaquin River between Hog and Turner Cuts (910)	0.136	0.007	0.108	0.008	NS
San Joaquin River, West of Oulton Point (812)	0.103	0.003	0.090	0.010	NS
Sacramento Deep Water Channel, Light 55	0.081	0.009	0.097	0.007	NS
Sacramento River at tip of Grand Island (711)	0.087	0.005	0.076	0.003	NS
Sacramento R. across from Sherman Lake (704)	0.096	0.004	0.109	0.006	NS
Middle of Broad Slough, West end (804)	0.070	0.005	0.072	0.004	NS
Field Dup.: Sacramento River at tip of Grand Island (711)	0.082	0.005	-	-	NA

	MSD	PMSD
One-way ANOVA	0.031	52.2
Two-way ANOVA	0.038	65.5

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

Table A46-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 10/31/06.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Old River, western arm at railroad bridge (902)	296	15.7	8.6	8.7	2.2	0.01	0.001
Old River at mouth of Holland Cut (915)	269	15.8	8.68	8.6	2.0	0.02	0.002
San Joaquin River between Hog and Turner Cuts (910)	339	15.0	8.28	7.9	6.0	0.11	0.005
San Joaquin River, West of Oulton Point (812)	258	15.8	8.43	8.4	3.7	0.08	0.005
Sacramento Deep Water Channel, Light 55	181	15.6	8.34	8.3	22.7	0.21	0.012
Sacramento River at tip of Grand Island (711)	156	15.1	8.26	8.4	4.8	0.29	0.013
Sacramento R. across from Sherman Lake (704)	4605	16.3	7.88	8.4	14.3	0.10	0.002
Middle of Broad Slough, West end (804)	5550	16.7	8.52	8.5	9.3	0.08	0.006
Field Dup.: Sacramento River at tip of Grand Island (711)	154	14.8	8.4	8.4	4.7	0.26	0.016

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Table A46-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 11/2/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 10/31/06.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	312	21.8	23.4	6.9	8.6	7.66	7.90	108	58	-
Old River, western arm at railroad bridge (902)	267	22.1	23.7	6.6	8.6	7.68	7.87	76	60	0.000
Old River at mouth of Holland Cut (915)	250	22.0	23.8	6.5	8.9	7.69	7.86	68	42	0.001
San Joaquin River between Hog and Turner Cuts (910)	316	22.3	23.8	6.6	8.7	7.66	7.86	80	64	0.002
San Joaquin River, West of Oulton Point (812)	233	21.8	23.5	6.7	8.9	7.61	7.88	68	62	0.003
Sacramento Deep Water Channel, Light 55	172	21.8	23.7	6.3	8.6	7.61	7.92	60	68	0.007
Sacramento River at tip of Grand Island (711)	155	21.6	23.7	6.3	8.7	7.55	7.91	52	60	0.010
Sacramento R. across from Sherman Lake (704)	4024	22.2	23.3	7.1	8.6	7.64	7.79	496	70	0.002
Middle of Broad Slough, West end (804)	5925	21.9	23.7	6.9	8.7	7.55	7.71	772	76	0.001
Field Dup.: Sacramento River at tip of Grand Island (711)	174	22.0	23.5	6.0	8.5	7.54	7.97	60	60	0.011
DIEPAMHR + 25 ppb PBO	322	22.1	23.4	7.1	8.5	7.72	8.11	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	261	22.2	23.2	6.9	8.7	7.39	7.85	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	249	22.3	23.2	7.0	8.7	7.41	7.89	-	-	-
San Joaquin River between Hog and Turner Cuts (910) + 25 ppb PBO	315	22.4	23.0	6.9	8.5	7.59	7.90	-	-	-
San Joaquin River, West of Oulton Point (812) + 25 ppb PBO	236	21.9	22.7	6.8	8.7	7.67	8.14	-	-	-
Sacramento Deep Water Channel, Light 55 + 25 ppb PBO	169	22.1	22.5	6.5	8.8	7.69	8.18	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	153	21.9	22.6	6.9	8.6	7.57	8.20	-	-	-
Sacramento R. across from Sherman Lake (704) + 25 ppb PBO	3981	21.8	22.5	7.0	8.7	7.59	7.95	-	-	-
Middle of Broad Slough, West end (804) + 25 ppb PBO	5620	21.5	22.7	7.0	8.8	7.57	7.91	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A47-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 11/03/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 11/01/06 - 11/02/06.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	92	4.1	37	12.0	S (40%)
High EC Control @ 15.42 mS/cm	90	6.2	97	3.3	NS
High EC Control @ 20.92 mS/cm	57	6.6	72	4.0	NS
Suisun Bay off Chipps Island (508)	98	2.0	100	0.0	NS
Suisun Bay, East of middle point (504)	96	2.6	93	3.3	NS
Carquinez Strait, West of Benicia army dock (405) ⁴	88	3.7	93	6.7	NS
Grizzly Bay at Dolphin (602) ³	98	2.0	93	6.7	NS
Montezuma Slough at Nurse Slough (609)	98	2.0	97	3.3	NS
Napa River at Riverside Blvd. terminus (340) ⁴	63	6.3	70	1.8	NS

	MSD	PMSD
One-way ANOVA	20.3	22.1
Two-way ANOVA	25.7	27.9

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.039	0.002	0.069	0.025	NS
High EC Control @ 15.42 mS/cm	0.036	0.003	0.036	0.003	NS
High EC Control @ 20.92 mS/cm	0.046	0.005	0.034	0.008	NS
Suisun Bay off Chipps Island (508)	0.070	0.005	0.061	0.007	NS
Suisun Bay, East of middle point (504)	0.060	0.007	0.061	0.005	NS
Carquinez Strait, West of Benicia army dock (405) ⁴	0.047	0.005	0.039	0.001	NS
Grizzly Bay at Dolphin (602) ³	0.058	0.002	0.047	0.004	NS
Montezuma Slough at Nurse Slough (609)	0.080	0.003	0.066	0.009	NS
Napa River at Riverside Blvd. terminus (340) ⁴	0.042	0.006	0.037	0.008	NS

	MSD	PMSD
One-way ANOVA	0.021	54.0
Two-way ANOVA	0.035	88.1

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. This high conductivity sample was compared to the High EC control @ 15.42 mS/cm.

4. These high conductivity samples were compared to the High EC control @ 20.92 mS/cm.

Table A47-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 11/01/06 - 11/02/06.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Suisun Bay off Chipps Island (508)	6550	15.1	7.81	8.9	7.1	0.11	0.001
Suisun Bay, East of middle point (504)	10390	15.3	7.73	9.0	5.3	0.09	0.001
Carquinez Strait, West of Benicia army dock (405)	22340	14.3	7.71	8.3	8.2	0.11	0.001
Grizzly Bay at Dolphin (602)	17830	15.0	7.81	8.7	11.7	0.09	0.001
Montezuma Slough at Nurse Slough (609)	10720	15.0	7.72	8.3	20.3	0.04	0.000
Napa River at Riverside Blvd. terminus (340)	24040	15.6	7.43	9.6	5.1	0.09	0.000

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Table A47-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 11/03/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 11/01/06 - 11/02/06.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	337	22.3	23.8	6.6	8.7	7.76	8.11	108	58	-
High EC Control @ 15.42 mS/cm	15260	22.4	24.5	6.8	8.4	7.57	7.72	1940	95	-
High EC Control @ 20.92 mS/cm	21050	22.1	24.6	7.1	8.7	7.66	7.87	2720	105	-
Suisun Bay off Chipps Island (508)	6125	22.5	24.5	6.6	8.6	7.56	7.84	692	74	0.002
Suisun Bay, East of middle point (504)	9830	22.5	24.4	6.8	8.8	7.65	7.82	210	83	0.002
Carquinez Strait, West of Benicia army dock (405)	20125	22.4	22.6	6.4	8.9	7.52	7.83	580	94	0.001
Grizzly Bay at Dolphin (602)	16275	22.3	23.6	6.8	8.6	7.61	7.86	362	100	0.001
Montezuma Slough at Nurse Slough (609)	9970	22.4	24.3	7.0	8.5	7.66	7.91	270	90	0.001
Napa River at Riverside Blvd. terminus (340)	22260	22.4	24.5	6.9	8.5	7.58	8.05	556	134	0.001
DIEPAMHR + 25 ppb PBO	457	22.2	23.5	7.1	8.5	7.91	8.30	-	-	-
High EC Control @ 15.42 mS/cm + 25 ppb PBO	15030	22.1	24.2	7.1	8.7	7.74	7.79	-	-	-
High EC Control @ 20.92 mS/cm + 25 ppb PBO	21440	22.2	24.2	7.0	8.4	7.64	7.86	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	6095	22.2	23.6	7.2	8.6	7.69	7.87	-	-	-
Suisun Bay, East of middle point (504) + 25 ppb PBO	9755	22.2	23.7	7.1	8.5	7.63	7.83	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	20960	22.3	23.9	7.0	8.5	7.57	7.89	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	13465	22.2	23.8	7.0	8.3	7.67	7.89	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	12895	22.2	23.6	6.8	8.5	7.59	7.97	-	-	-
Napa River at Riverside Blvd. terminus (340) + 25 ppb PBO	21715	22.2	23.8	7.3	8.6	7.61	7.98	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A48-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 11/16/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 11/14/06.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO ²
DIEPAMHR	92	2.6	98	2.5	NS
DIEPAMHR + organic matter	100	0.0	-	-	NA
Old River, western arm at railroad bridge (902)	95	5.0	100	0.0	NS
Old River at mouth of Holland Cut (915)	95	2.9	93	2.4	NS
San Joaquin River between Hog and Turner Cuts (910)	98	2.5	100	0.0	NS
San Joaquin River, West of Oulton Point (812)	98	2.5	100	0.0	NS
Sacramento River at tip of Grand Island (711)	95	2.9	84	7.1	NS
Sacramento Deep Water Channel, Light 55	90	0.4	92	4.8	NS
Sacramento R. across from Sherman Lake (704)	100	0.0	95	5.0	NS
Middle of Broad Slough, West end (804)	100	0.0	94	5.6	NS

	MSD	PMSD
One-way ANOVA	12.1	12.1
Two-way ANOVA	17.6	17.6

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO ²
DIEPAMHR	0.067	0.006	0.054	0.006	NS
DIEPAMHR + organic matter	0.077	0.004	-	-	NA
Old River, western arm at railroad bridge (902)	0.093	0.004	0.079	0.008	NS
Old River at mouth of Holland Cut (915)	0.083	0.007	0.080	0.009	NS
San Joaquin River between Hog and Turner Cuts (910)	0.084	0.005	0.061	0.011	NS
San Joaquin River, West of Oulton Point (812)	0.085	0.002	0.081	0.008	NS
Sacramento River at tip of Grand Island (711)	0.063	0.005	0.069	0.007	NS
Sacramento Deep Water Channel, Light 55	0.076	0.008	0.065	0.003	NS
Sacramento R. across from Sherman Lake (704)	0.070	0.009	0.064	0.008	NS
Middle of Broad Slough, West end (804)	0.075	0.007	0.064	0.008	NS

	MSD	PMSD
One-way ANOVA	0.028	36.2
Two-way ANOVA	0.037	47.5

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

Table A48-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 11/14/06.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Old River, western arm at railroad bridge (902)	388	14.4	8.06	10.1	2.5	0.03	0.001
Old River at mouth of Holland Cut (915)	409	14.2	7.9	10.2	2.6	0.01	0.000
San Joaquin River between Hog and Turner Cuts (910)	519	14.6	7.74	9.4	7.5	0.06	0.001
San Joaquin River, West of Oulton Point (812)	576	14.6	7.87	10.2	7.4	0.06	0.001
Sacramento River at tip of Grand Island (711)	174	14.2	7.75	9.2	6.6	0.31	0.004
Sacramento Deep Water Channel, Light 55	179	14.1	7.79	9.7	11.6	0.26	0.004
Sacramento R. across from Sherman Lake (704)	5540	15.2	8	10.0	12.9	0.10	0.002
Middle of Broad Slough, West end (804)	5440	15.0	7.95	10.0	10.7	0.08	0.001

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Table A48-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 11/16/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 11/14/06.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	362	23.3	24.0	7.6	8.9	7.75	8.15	100	60	-
DIEPAMHR + organic matter	364	22.9	23.4	7.7	8.7	7.75	8.15	100	60	-
Old River, western arm at railroad bridge (902)	422	23.4	23.9	7.5	8.4	7.75	8.10	78	68	0.002
Old River at mouth of Holland Cut (915)	407	23.3	23.8	7.6	8.3	7.76	8.19	80	63	0.001
San Joaquin River between Hog and Turner Cuts (910)	545	23.3	23.6	7.5	8.3	7.80	8.10	124	95	0.003
San Joaquin River, West of Oulton Point (812)	557	23.3	24.0	7.6	8.8	7.71	8.03	92	60	0.003
Sacramento River at tip of Grand Island (711)	193	23.2	24.1	7.3	8.5	7.70	8.10	62	68	0.018
Sacramento Deep Water Channel, Light 55	198	23.2	23.9	7.6	8.4	7.74	8.13	64	71	0.016
Sacramento R. across from Sherman Lake (704)	5780	23.0	23.9	7.8	8.3	7.73	7.97	618	69	0.004
Middle of Broad Slough, West end (804)	5515	23.2	23.8	7.8	8.4	7.71	8.04	606	70	0.003
DIEPAMHR + 25 ppb PBO	388	23.3	24.0	7.6	8.7	7.74	8.21	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	424	23.4	23.9	7.8	8.6	7.71	8.18	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	433	23.2	23.7	7.3	8.4	7.69	8.14	-	-	-
San Joaquin River between Hog and Turner Cuts (910) + 25 ppb PBO	547	23.3	23.7	7.5	8.7	7.81	8.10	-	-	-
San Joaquin River, West of Oulton Point (812) + 25 ppb PBO	552	23.4	23.6	7.8	8.6	7.78	8.09	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	201	23.3	24.1	7.3	8.6	7.68	8.08	-	-	-
Sacramento Deep Water Channel, Light 55 + 25 ppb PBO	202	23.2	24.0	7.4	8.9	7.75	8.04	-	-	-
Sacramento R. across from Sherman Lake (704) + 25 ppb PBO	5715	23.2	23.8	7.9	8.5	7.62	7.91	-	-	-
Middle of Broad Slough, West end (804) + 25 ppb PBO	5355	23.2	23.7	7.8	8.7	7.71	7.93	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A49-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 11/17/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 11/15/06 - 11/16/06.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	89	4.5	98	2.5	NS
High EC Control @ 12.0 mS/cm	95	2.9	100	0.0	NS
High EC Control @ 17.0 mS/cm	70	23.5	90	4.1	NS
High EC Control @ 22.0 mS/cm	81	4.6	84	6.9	NS
Suisun Bay off Chipps Island (508)	98	2.5	95	2.9	NS
Suisun Bay, East of middle point (504) ³	100	0.0	98	2.5	NS
Carquinez Strait, West of Benicia army dock (405) ⁵	83	10.8	81	4.1	NS
Grizzly Bay at Dolphin (602) ⁴	98	2.5	95	5.0	NS
Montezuma Slough at Nurse Slough (609) ³	100	0.0	95	2.9	NS
Napa River at Riverside Blvd. terminus (340) ⁴	95	5.0	92	4.8	NS
Trip Blank	93	7.5	-	-	NA

	MSD	PMSD
One-way ANOVA	42.1	47.0
Two-way ANOVA	22.7	25.4

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.052	0.013	0.060	0.012	NS
High EC Control @ 12.0 mS/cm	0.035	0.013	0.050	0.015	NS
High EC Control @ 17.0 mS/cm	0.047	0.009	0.057	0.013	NS
High EC Control @ 22.0 mS/cm	0.049	0.010	0.066	0.018	NS
Suisun Bay off Chipps Island (508)	0.085	0.005	0.059	0.011	NS
Suisun Bay, East of middle point (504) ³	0.083	0.020	0.064	0.007	NS
Carquinez Strait, West of Benicia army dock (405) ⁵	0.017	0.004	0.042	0.007	NS
Grizzly Bay at Dolphin (602) ⁴	0.053	0.011	0.039	0.004	NS
Montezuma Slough at Nurse Slough (609) ³	0.069	0.013	0.064	0.008	NS
Napa River at Riverside Blvd. terminus (340) ⁴	0.052	0.009	0.040	0.004	NS
Trip Blank	0.056	0.003	-	-	NA

	MSD	PMSD
One-way ANOVA	0.054	105.6
Two-way ANOVA	0.059	114.8

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. These high conductivity samples were compared to the High EC control @ 12.0 mS/cm.

4. These high conductivity samples were compared to the High EC control @ 17.0 mS/cm.

5. This high conductivity sample was compared to the High EC control @ 22.0 mS/cm.

Table A49-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 11/15/06 - 11/16/06.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Suisun Bay off Chipps Island (508)	7910	14.4	7.83	10.3	8.0	0.08	0.001
Suisun Bay, East of middle point (504)	12360	14.5	7.86	10.0	8.0	0.09	0.001
Carquinez Strait, West of Benicia army dock (405)	22140	14.8	7.83	9.6	11.8	0.15	0.002
Grizzly Bay at Dolphin (602)	17280	14.5	7.91	9.9	20.5	0.07	0.001
Montezuma Slough at Nurse Slough (609)	11910	14.3	7.67	9.6	27.9	0.09	0.001
Napa River at Riverside Blvd. terminus (340)	18280	14.3	7.41	8.8	4.9	0.02	0.000
Trip Blank	423	14.5	8.38	10.7	0.3	0.00	0.000

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Table A49-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 11/17/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 11/15/06 - 11/16/06.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	372	23.1	24.6	7.5	8.6	7.88	8.16	100	60	-
High EC Control @ 12.0 mS/cm	12475	23.4	24.4	7.8	8.6	7.71	7.81	1380	85	-
High EC Control @ 17.0 mS/cm	17635	23.4	24.5	7.9	8.5	7.74	7.86	2040	95	-
High EC Control @ 22.0 mS/cm	22495	23.4	24.3	7.9	8.3	7.78	7.86	2560	110	-
Suisun Bay off Chipps Island (508)	8465	23.4	24.3	7.7	8.8	7.77	7.95	920	75	0.003
Suisun Bay, East of middle point (504)	12725	23.3	24.5	7.6	8.8	7.72	7.85	1380	90	0.002
Carquinez Strait, West of Benicia army dock (405)	23075	23.2	24.3	7.5	8.7	7.69	7.83	2680	110	0.003
Grizzly Bay at Dolphin (602)	18170	23.2	24.3	7.7	8.5	7.75	7.86	2000	100	0.001
Montezuma Slough at Nurse Slough (609)	12570	23.1	24.3	7.6	8.9	7.64	8.00	1410	94	0.001
Napa River at Riverside Blvd. terminus (340)	17950	23.1	24.2	7.6	8.6	7.58	8.06	140	124	0.000
Trip Blank	487	22.9	24.1	7.7	8.4	7.88	8.22	108	58	0.000
DIEPAMHR	470	23.1	23.6	7.7	8.3	7.90	8.23	-	-	-
High EC Control @ 12.0 mS/cm	12400	23.0	23.6	7.9	8.6	7.77	7.87	-	-	-
High EC Control @ 17.0 mS/cm	17865	23.1	23.6	7.9	8.4	7.74	7.90	-	-	-
High EC Control @ 22.0 mS/cm	23025	22.9	23.7	8.0	8.6	7.78	7.91	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	8395	22.8	23.8	7.8	8.7	7.80	7.98	-	-	-
Suisun Bay, East of middle point (504) + 25 ppb PBO	12350	22.8	23.9	7.5	8.7	7.73	7.92	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	23025	22.8	23.9	7.7	8.9	7.76	7.88	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	17710	22.8	24.2	7.6	8.7	7.75	7.89	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	12420	22.8	24.2	7.9	8.9	7.72	7.99	-	-	-
Napa River at Riverside Blvd. terminus (340) + 25 ppb PBO	17630	22.7	24.2	7.8	8.5	7.75	8.08	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A50-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 11/30/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 11/28/06.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	98	2.5	98	2.5	NS
Old River, western arm at railroad bridge (902)	100	0.0	95	2.9	NS
Old River at mouth of Holland Cut (915)	100	0.0	100	0.0	NS
San Joaquin River between Hog and Turner Cuts (910)	100	0.0	100	0.0	NS
San Joaquin River, West of Oulton Point (812)	100	0.0	98	2.5	NS
Sacramento River at tip of Grand Island (711)	81	16.0	83	5.9	NS
Sacramento Deep Water Channel, Light 55	93	7.5	95	2.9	NS
Sacramento R. across from Sherman Lake (704)	98	2.5	95	2.8	NS
Middle of Broad Slough, West end (804)	100	0.0	98	2.5	NS

	MSD	PMSD
One-way ANOVA	28.6	29.3
Two-way ANOVA	24.6	25.2

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.062	0.006	0.047	0.008	NS
Old River, western arm at railroad bridge (902)	0.105	0.008	0.082	0.013	NS
Old River at mouth of Holland Cut (915)	0.095	0.008	0.107	0.008	NS
San Joaquin River between Hog and Turner Cuts (910)	0.087	0.006	0.100	0.010	NS
San Joaquin River, West of Oulton Point (812)	0.083	0.007	0.097	0.011	NS
Sacramento River at tip of Grand Island (711)	0.050	0.008	0.073	0.013	NS
Sacramento Deep Water Channel, Light 55	0.079	0.010	0.092	0.008	NS
Sacramento R. across from Sherman Lake (704)	0.097	0.011	0.107	0.012	NS
Middle of Broad Slough, West end (804)	0.102	0.007	0.091	0.006	NS

	MSD	PMSD
One-way ANOVA	0.038	60.3
Two-way ANOVA	0.041	65.9

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

Table A50-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 11/28/07.

Treatment	Field Chemistry				Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)		
Old River, western arm at railroad bridge (902)	556	11.5	7.64	10.5	0.08	0.001
Old River at mouth of Holland Cut (915)	461	12.2	7.35	10.3	0.07	0.000
San Joaquin River between Hog and Turner Cuts (910)	426	12.1	7.48	9.2	0.14	0.001
San Joaquin River, West of Oulton Point (812)	695	12.8	7.58	9.9	0.15	0.001
Sacramento River at tip of Grand Island (711)	695	12.8	7.58	9.9	0.38	0.003
Sacramento Deep Water Channel, Light 55	218	11.7	7.60	9.7	0.25	0.002
Sacramento R. across from Sherman Lake (704)	2381	12.7	7.40	9.9	0.19	0.001
Middle of Broad Slough, West end (804)	4674	12.6	7.50	9.9	0.19	0.001

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Table A50-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 11/30/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 11/28/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	457	22.1	22.9	7.3	8.9	7.93	8.11	106	61	-
Old River, western arm at railroad bridge (902)	585	22.8	23.1	7.0	8.8	7.85	8.03	96	70	0.004
Old River at mouth of Holland Cut (915)	488	22.6	22.7	7.0	8.9	7.84	8.08	84	72	0.003
San Joaquin River between Hog and Turner Cuts (910)	472	22.2	22.3	7.1	8.8	7.87	8.13	108	92	0.008
San Joaquin River, West of Oulton Point (812)	663	23.2	23.3	6.9	8.8	7.81	8.01	112	82	0.007
Sacramento River at tip of Grand Island (711)	211	22.9	23.2	7.1	8.9	7.80	8.06	68	78	0.019
Sacramento Deep Water Channel, Light 55	238	22.7	22.9	7.2	8.8	7.82	8.17	80	80	0.016
Sacramento R. across from Sherman Lake (704)	4909	22.5	22.8	7.2	8.7	7.76	7.90	496	72	0.006
Middle of Broad Slough, West end (804)	4708	22.5	23.1	7.3	8.8	7.81	7.88	504	80	0.005
DIEPAMHR + 25 ppb PBO	395	22.0	23.3	7.3	8.8	7.93	8.22	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	604	22.1	23.2	7.2	8.9	7.89	8.14	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	478	22.4	23.3	7.1	8.8	7.89	8.17	-	-	-
San Joaquin River between Hog and Turner Cuts (910) + 25 ppb PBO	453	22.7	23.3	7.2	8.8	7.93	8.16	-	-	-
San Joaquin River, West of Oulton Point (812) + 25 ppb PBO	751	22.3	23.3	7.0	8.9	7.84	8.07	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	217	22.4	23.3	7.1	8.7	7.88	8.12	-	-	-
Sacramento Deep Water Channel, Light 55 + 25 ppb PBO	248	23.1	23.4	6.9	8.9	7.90	8.14	-	-	-
Sacramento R. across from Sherman Lake (704) + 25 ppb PBO	4908	22.9	23.2	7.2	8.7	7.81	7.89	-	-	-
Middle of Broad Slough, West end (804) + 25 ppb PBO	4761	22.6	23.3	7.3	8.8	7.85	7.93	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A51-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 12/01/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 11/29/06 - 11/30/06.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	100	0.0	98	2.5	NS
High EC control @ 12.0 mS/cm	98	2.5	100	0.0	NS
High EC control @ 16.5 mS/cm	98	2.5	98	2.5	NS
High EC control @ 22.0 mS/cm	88	5.1	79	3.9	NS
Suisun Bay off Chipps Island (508)	100	0.0	100	0.0	NS
Suisun Bay, East of middle point (504) ³	100	0.0	98	2.5	NS
Carquinez Strait, West of Benicia army dock (405) ⁵	93	2.5	82	2.2	NS
Grizzly Bay at Dolphin (602) ⁴	100	0.0	98	2.5	NS
Montezuma Slough at Nurse Slough (609) ³	98	2.5	100	0.0	NS
Napa River at Riverside Blvd. terminus (340) ⁴	100	0.0	98	2.5	NS

	MSD	PMSD
One-way ANOVA	10.9	10.9
Two-way ANOVA	11.9	11.9

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.060	0.004	0.061	0.002	NS
High EC control @ 12.0 mS/cm	0.073	0.005	0.057	0.004	NS
High EC control @ 16.5 mS/cm	0.053	0.003	0.054	0.005	NS
High EC control @ 22.0 mS/cm	0.045	0.005	0.044	0.002	NS
Suisun Bay off Chipps Island (508)	0.097	0.007	0.108	0.003	NS
Suisun Bay, East of middle point (504) ³	0.084	0.007	0.079	0.002	NS
Carquinez Strait, West of Benicia army dock (405) ⁵	0.047	0.003	0.053	0.007	NS
Grizzly Bay at Dolphin (602) ⁴	0.062	0.002	0.065	0.004	NS
Montezuma Slough at Nurse Slough (609) ³	0.089	0.010	0.079	0.003	NS
Napa River at Riverside Blvd. terminus (340) ⁴	0.053	0.001	0.052	0.002	NS

	MSD	PMSD
One-way ANOVA	0.026	44.2
Two-way ANOVA	0.024	40.8

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. These high conductivity samples were compared to the High EC control @ 12.0 mS/cm.

4. These high conductivity samples were compared to the High EC control @ 16.5 mS/cm.

5. This high conductivity sample was compared to the High EC control @ 22.0 mS/cm.

Table A51-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 11/29/06 - 11/30/06.

Treatment	Field Chemistry				Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)		
Suisun Bay off Chipps Island (508)	8240	11.5	7.52	10.1	0.17	0.001
Suisun Bay, East of middle point (504)	10510	11.4	7.53	10.0	0.14	0.001
Carquinez Strait, West of Benicia army dock (405)	21680	11.5	7.76	9.4	0.18	0.001
Grizzly Bay at Dolphin (602)	16620	11.4	7.77	9.9	0.14	0.001
Montezuma Slough at Nurse Slough (609)	12460	11.8	7.39	8.3	0.25	0.001
Napa River at Riverside Blvd. terminus (340)	16750	12.0	7.86	11.6	0.00	0.000

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Table A51-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 12/01/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 11/29/06 - 11/30/06.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	380	23.1	23.5	7.0	8.8	7.87	8.15	106	61	-
High EC control @ 12.0 mS/cm	12645	23.2	24.2	7.2	8.8	7.67	7.78	1460	90	-
High EC control @ 16.5 mS/cm	17990	23.8	24.3	7.2	8.5	7.71	7.85	2080	100	-
High EC control @ 22.0 mS/cm	22830	23.9	24.0	7.3	8.7	7.68	7.84	2600	100	-
Suisun Bay off Chipps Island (508)	8630	23.8	23.9	7.0	8.6	7.76	7.88	940	82	0.005
Suisun Bay, East of middle point (504)	11005	23.6	24.0	7.2	8.7	7.73	7.86	1360	100	0.004
Carquinez Strait, West of Benicia army dock (405)	24010	23.8	24.3	6.9	8.7	7.44	7.76	2720	110	0.003
Grizzly Bay at Dolphin (602)	16645	23.9	24.0	7.4	8.6	7.76	7.91	2080	110	0.004
Montezuma Slough at Nurse Slough (609)	12645	23.5	23.8	7.3	8.4	7.79	7.93	1600	120	0.005
Napa River at Riverside Blvd. terminus (340)	17180	23.8	24.4	7.7	8.9	7.77	7.94	2060	140	0.000
DIEPAMHR + 25 ppb PBO	385	23.3	23.6	6.9	8.5	7.92	8.20	-	-	-
High EC control @ 12.0 mS/cm + 25 ppb PBO	12785	23.8	24.1	7.2	8.7	7.70	7.82	-	-	-
High EC control @ 16.5 mS/cm + 25 ppb PBO	17565	23.7	24.0	7.0	8.4	7.70	7.86	-	-	-
High EC control @ 22.0 mS/cm + 25 ppb PBO	22635	23.7	24.1	7.3	8.7	7.70	7.85	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	8830	23.5	23.9	7.0	8.6	7.76	7.89	-	-	-
Suisun Bay, East of middle point (504) + 25 ppb PBO	10895	22.6	24.0	7.4	8.4	7.73	7.81	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	22475	23.8	24.3	7.6	8.6	7.69	7.79	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	17605	23.6	24.4	7.5	8.4	7.78	7.82	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	12710	23.6	24.2	7.5	8.2	7.81	7.85	-	-	-
Napa River at Riverside Blvd. terminus (340) + 25 ppb PBO	17145	23.5	24.3	7.5	8.6	7.78	7.95	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A52-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 12/13/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 12/12/06.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	100	0.0	90	10.0	NS
Old River, western arm at railroad bridge (902)	100	0.0	100	0.0	NS
Old River at mouth of Holland Cut (915)	98	2.5	100	0.0	NS
San Joaquin River between Hog and Turner Cuts (910)	100	0.0	100	0.0	NS
San Joaquin River, West of Oulton Point (812)	98	2.3	100	0.0	NS
Sacramento River at tip of Grand Island (711)	93	4.8	89	4.5	NS
Sacramento Deep Water Channel, Light 55	98	2.5	100	0.0	NS
Sacramento R. across from Sherman Lake (704)	100	0.0	98	2.5	NS
Middle of Broad Slough, West end (804)	100	0.0	100	0.0	NS

	MSD	PMSD
One-way ANOVA	10.2	10.2
Two-way ANOVA	15.9	15.9

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.076	0.004	0.062	0.004	NS
Old River, western arm at railroad bridge (902)	0.117	0.008	0.103	0.007	NS
Old River at mouth of Holland Cut (915)	0.123	0.003	0.113	0.018	NS
San Joaquin River between Hog and Turner Cuts (910)	0.112	0.004	0.099	0.004	NS
San Joaquin River, West of Oulton Point (812)	0.109	0.011	0.097	0.006	NS
Sacramento River at tip of Grand Island (711)	0.073	0.006	0.062	0.009	NS
Sacramento Deep Water Channel, Light 55	0.100	0.008	0.075	0.007	NS
Sacramento R. across from Sherman Lake (704)	0.100	0.006	0.084	0.007	NS
Middle of Broad Slough, West end (804)	0.131	0.004	0.103	0.004	NS

	MSD	PMSD
One-way ANOVA	0.032	41.3
Two-way ANOVA	0.039	51.3

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

Table A52-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 12/12/06.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Old River, western arm at railroad bridge (902)	723	10.7	7.49	11.6	2.6	0.08	0.000
Old River at mouth of Holland Cut (915)	645	10.6	7.18	12.1	2.9	0.08	0.000
San Joaquin River between Hog and Turner Cuts (910)	297	10.3	7.15	10.9	4.5	0.13	0.000
San Joaquin River, West of Oulton Point (812)	811	10.4	7.41	11.3	4.3	0.13	0.001
Sacramento River at tip of Grand Island (711)	191	10.2	7.41	11.2	4.7	0.34	0.002
Sacramento Deep Water Channel, Light 55	269	10.1	7.49	11.2	9.5	0.19	0.001
Sacramento R. across from Sherman Lake (704)	1833	10.2	7.3	11.1	8.1	0.20	0.001
Middle of Broad Slough, West end (804)	3821	10.7	7.33	11.2	6.3	0.14	0.000

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Table A52-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 12/13/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 12/12/06.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	382	22.7	24.7	7.7	8.6	7.91	8.08	104	59	-
Old River, western arm at railroad bridge (902)	755	22.3	24.7	7.6	8.6	7.95	8.08	118	72	0.005
Old River at mouth of Holland Cut (915)	606	22.5	24.4	7.7	8.4	7.88	8.07	104	71	0.004
San Joaquin River between Hog and Turner Cuts (910)	320	22.2	24.5	7.6	8.6	7.89	8.13	82	74	0.008
San Joaquin River, West of Oulton Point (812)	770	22.1	24.5	7.6	8.7	7.85	8.06	118	73	0.007
Sacramento River at tip of Grand Island (711)	205	21.9	24.5	7.2	8.7	7.83	8.13	72	76	0.022
Sacramento Deep Water Channel, Light 55	290	21.9	24.3	7.5	8.8	7.97	8.14	92	100	0.012
Sacramento R. across from Sherman Lake (704)	1896	21.5	24.4	7.6	8.7	7.83	7.98	224	77	0.008
Middle of Broad Slough, West end (804)	3822	21.5	25.5	7.8	8.6	7.77	8.04	396	77	0.007
DIEPAMHR + 25 ppb PBO	376	21.7	24.0	7.7	8.7	7.82	8.11	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	752	21.5	24.2	7.7	8.7	7.93	8.08	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	603	21.4	24.9	7.6	8.8	7.83	8.13	-	-	-
San Joaquin River between Hog and Turner Cuts (910) + 25 ppb PBO	348	21.4	25.1	7.7	8.4	7.86	8.08	-	-	-
San Joaquin River, West of Oulton Point (812) + 25 ppb PBO	775	21.3	25.3	7.7	8.9	7.81	7.97	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	213	21.2	24.8	7.5	8.4	7.85	8.14	-	-	-
Sacramento Deep Water Channel, Light 55 + 25 ppb PBO	290	20.8	24.9	7.7	8.8	7.94	8.13	-	-	-
Sacramento R. across from Sherman Lake (704) + 25 ppb PBO	1888	20.6	24.8	7.8	8.8	7.82	8.06	-	-	-
Middle of Broad Slough, West end (804) + 25 ppb PBO	3769	20.6	24.7	7.9	8.9	7.80	8.00	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A53-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 12/14/06 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 12/13/06.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	100	0.0	100	0.0	NS
High EC Control @ 13.0 mS/cm	100	0.0	100	0.0	NS
High EC Control @ 18.0 mS/cm	98	2.5	100	0.0	NS
High EC Control @ 21.5 mS/cm	98	2.5	97	2.8	NS
Suisun Bay off Chipps Island (508)	100	0.0	100	0.0	NS
Suisun Bay, East of middle point (504) ⁴	100	0.0	100	0.0	NS
Carquinez Strait, West of Benicia army dock (405) ⁵	98	2.5	95	2.9	NS
Grizzly Bay at Dolphin (602) ⁵	98	2.5	98	2.5	NS
Montezuma Slough at Nurse Slough (609) ³	100	0.0	95	2.9	NS
Napa River at Riverside Blvd. terminus (340)	100	0.0	100	0.0	NS

	MSD	PMSD
One-way ANOVA	7.6	7.6
Two-way ANOVA	8.8	8.8

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.066	0.018	0.066	0.013	NS
High EC Control @ 13.0 mS/cm	0.066	0.006	0.051	0.002	NS
High EC Control @ 18.0 mS/cm	0.066	0.006	0.033	0.006	NS
High EC Control @ 21.5 mS/cm	0.055	0.006	0.034	0.007	NS
Suisun Bay off Chipps Island (508)	0.116	0.015	0.086	0.016	NS
Suisun Bay, East of middle point (504) ⁴	0.083	0.005	0.054	0.015	NS
Carquinez Strait, West of Benicia army dock (405) ⁵	0.070	0.007	0.032	0.007	NS
Grizzly Bay at Dolphin (602) ⁵	0.071	0.012	0.059	0.003	NS
Montezuma Slough at Nurse Slough (609) ³	0.092	0.009	0.074	0.008	NS
Napa River at Riverside Blvd. terminus (340)	0.105	0.010	0.102	0.009	NS

	MSD	PMSD
One-way ANOVA	0.049	75.1
Two-way ANOVA	0.052	79.5

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using USEPA standard single-sample statistical protocols modified for a multiple sample design.

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. This high conductivity sample was compared to the High EC control @ 13.0 mS/cm.

4. This high conductivity sample was compared to the High EC control @ 18.0 mS/cm.

5. These high conductivity samples were compared to the High EC control @ 21.5 mS/cm.

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Table A53-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 2/13/06.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Suisun Bay off Chipps Island (508)	9390	11.0	7.70	11.3	8.1	0.17	0.001
Suisun Bay, East of middle point (504)	17540	11.3	7.79	10.9	11.9	0.17	0.001
Carquinez Strait, West of Benicia army dock (405)	20400	11.5	7.86	10.9	6.9	0.16	0.002
Grizzly Bay at Dolphin (602)	18860	11.6	7.84	10.5	7.9	0.15	0.001
Montezuma Slough at Nurse Slough (609)	12970	11.4	7.48	9.6	17.5	0.20	0.001
Napa River at Riverside Blvd. terminus (340)	6030	12.4	7.65	10.9	10.8	0.05	0.000

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Table A53-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 12/14/06 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 12/13/06.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	369	23.0	23.7	7.5	8.2	8.00	8.12	104	59	-
High EC Control @ 13.0 mS/cm	13005	23.2	23.6	7.6	8.7	7.70	7.79	1560	110	-
High EC Control @ 18.0 mS/cm	17290	23.3	24.1	7.4	8.4	7.69	7.79	2280	110	-
High EC Control @ 21.5 mS/cm	22380	23.3	23.7	7.3	8.5	7.69	7.84	2500	110	-
Suisun Bay off Chipps Island (508)	9165	23.1	24.5	7.4	8.8	7.77	7.84	1000	82	0.004
Suisun Bay, East of middle point (504)	17870	23.2	23.6	7.3	8.8	7.67	7.83	1920	89	0.003
Carquinez Strait, West of Benicia army dock (405)	20240	23.2	24.8	6.7	8.4	7.66	7.71	2270	92	0.003
Grizzly Bay at Dolphin (602)	19280	23.2	24.3	7.1	8.4	7.70	7.78	2090	91	0.003
Montezuma Slough at Nurse Slough (609)	13420	23.1	24.8	7.5	8.2	7.80	7.91	1480	103	0.005
Napa River at Riverside Blvd. terminus (340)	5995	23.2	24.9	7.6	8.8	7.80	7.99	690	97	0.001
DIEPAMHR + 25 ppb PBO	383	22.8	24.6	7.4	8.6	7.82	8.16	-	-	-
High EC Control @ 13.0 mS/cm + 25 ppb PBO	13015	22.7	24.4	7.7	8.9	7.62	7.85	-	-	-
High EC Control @ 18.0 mS/cm + 25 ppb PBO	18065	23.2	24.5	7.5	8.9	7.67	7.80	-	-	-
High EC Control @ 21.5 mS/cm + 25 ppb PBO	22050	22.9	24.3	7.4	8.5	7.66	7.81	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	9325	22.7	24.3	7.6	8.8	7.79	7.92	-	-	-
Suisun Bay, East of middle point (504) + 25 ppb PBO	17835	23.0	24.3	7.2	8.7	7.66	7.81	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	20685	23.0	24.1	7.3	8.5	7.66	7.84	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	18945	22.7	24.4	7.1	8.7	7.69	7.84	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	13205	23.4	24.4	7.5	8.2	7.78	7.93	-	-	-
Napa River at Riverside Blvd. terminus (340) + 25 ppb PBO	6080	23.4	24.2	7.5	8.7	7.91	8.03	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A54-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 1/04/07 examining samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 1/02/07 - 1/03/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
Lab Control (DIEPAMHR)	100	0.0	-	-	NA
DIEPAMHR + organic matter	100	0.0	100	0.0	NS
High EC Control @ 12.0 mS/cm	100	0.0	95	2.8	NS
Middle of Broad Slough (804)	100	0.0	100	0.0	NS
Suisun Bay, off Chipps Island (508)	100	0.0	100	0.0	NS
Suisun Bay, East of middle point (504)	100	0.0	100	0.0	NS
Napa River @ Riverside Blvd Term. (340)	100	0.0	95	5.0	NS
Grizzly Bay @ Dolphin (602) ³	98	2.3	95	5.0	NS
Montezuma Slough @ Nurse Slough (609) ³	98	2.5	100	0.0	NS
Sacramento River N. side, across from Sherman L. (704)	100	0.0	100	0.0	NS

	MSD	PMSD
One-way ANOVA	5.2	5.2
Two-way ANOVA	10.2	10.2

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
Lab Control (DIEPAMHR)	0.062	0.007	-	-	NA
DIEPAMHR + organic matter	0.068	0.008	0.071	0.007	NS
High EC Control @ 12.0 mS/cm	0.035	0.008	0.048	0.002	NS
Middle of Broad Slough (804)	0.088	0.005	0.082	0.003	NS
Suisun Bay, off Chipps Island (508)	0.091	0.002	0.091	0.005	NS
Suisun Bay, East of middle point (504)	0.081	0.014	0.090	0.009	NS
Napa River @ Riverside Blvd Term. (340)	0.092	0.008	0.099	0.006	NS
Grizzly Bay @ Dolphin (602) ³	0.058	0.009	0.073	0.005	NS
Montezuma Slough @ Nurse Slough (609) ³	0.064	0.009	0.058	0.007	NS
Sacramento River N. side, across from Sherman L. (704)	0.108	0.011	0.124	0.011	NS

	MSD	PMSD
One-way ANOVA	0.041	60.9
Two-way ANOVA	0.041	59.9

1. Highlighted areas indicate a significant reduction in survival or weight compared to the nutrient addback or High EC control. Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$). Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
2. NS: Nonsignificant, S: Significant, NA: Not applicable.
3. These high conductivity samples were compared to the High EC Control.

Table A54-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 1/02/07 - 1/03/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Middle of Broad Slough (804)	1240	8.1	7.66	12.1	15.8	0.16	0.001
Suisun Bay, off Chipps Island (508)	3952	8.6	7.45	12.9	13.8	0.19	0.001
Suisun Bay, East of middle point (504)	7580	8.9	7.82	12.1	15.7	0.15	0.001
Napa River @ Riverside Blvd Term. (340)	4826	8.6	7.42	11.8	14.5	0.23	0.001
Grizzly Bay @ Dolphin (602)	11990	9.3	7.46	11.8	22.8	0.17	0.001
Montezuma Slough @ Nurse Slough (609)	12230	8.3	7.48	10.7	34.5	0.25	0.001
Sacramento River N. side, across from Sherman L. (704)	1058	8.6	7.52	11.4	35.3	0.22	0.001

Table A54-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 1/04/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 1/02/07 - 1/03/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	371	21.6	22.4	7.5	8.9	8.03	8.34	92	60	-
DIEPAMHR + nutrient	417	22.7	22.8	7.4	8.7	7.92	8.08	-	-	-
High EC Control @ 12.0 mS	11945	22.3	22.9	7.5	8.8	7.85	7.99	260	14	-
Middle of Broad Slough (804)	1285	23.3	23.3	7.7	8.8	8.00	8.14	172	74	0.007
Suisun Bay, off Chipps Island (508)	3701	22.7	23.3	7.3	8.5	7.85	7.97	408	78	0.006
Suisun Bay, East of middle point (504)	8230	23.2	23.2	7.7	8.2	7.86	8.00	840	100	0.005
Napa River @ Riverside Blvd Term. (340)	4958	23.1	23.3	7.5	8.4	7.94	8.01	536	92	0.008
Grizzly Bay @ Dolphin (602)	13060	22.7	22.8	7.6	8.9	7.81	7.97	1320	100	0.004
Montezuma Slough @ Nurse Slough (609)	12495	22.4	22.8	7.6	8.9	7.70	8.03	1380	110	0.004
Sacramento River N. side, across from Sherman L. (704)	909	22.7	22.8	7.6	8.9	7.99	8.19	124	72	0.009
DIEPAMHR + 1% nutrient addback + 25 ppb PBO	390	22.0	23.3	7.2	8.6	7.92	8.09	-	-	-
High EC Control @ 12.0 mS + 25 ppb PBO	12025	21.8	23.6	7.6	8.4	7.76	8.01	-	-	-
Middle of Broad Slough (804) + 25 ppb PBO	1202	22.0	22.6	7.5	8.6	7.98	8.08	-	-	-
Suisun Bay, off Chipps Island (508) + 25 ppb PBO	3537	22.0	22.6	7.4	8.7	7.94	8.03	-	-	-
Suisun Bay, East of middle point (504) + 25 ppb PBO	8050	21.9	22.6	7.8	8.4	7.80	8.04	-	-	-
Napa River @ Riverside Blvd Term. (340) + 25 ppb PBO	5016	22.0	22.5	7.4	8.9	7.90	8.04	-	-	-
Grizzly Bay @ Dolphin (602) + 25 ppb PBO	12875	21.9	22.6	7.5	8.6	7.78	8.02	-	-	-
Montezuma Slough @ Nurse Slough (609) + 25 ppb PBO	12600	21.8	23.4	7.2	8.4	7.77	8.05	-	-	-
Sacramento River N. side, across from Sherman L. (704) + 25 ppb PBO	823	21.7	22.6	7.0	8.7	7.91	8.18	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured a test initiation.

Table A55-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 1/05/2007 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 1/03/2007- 1/04/2007.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	98	2.5	-	-	NA
DIEPAMHR + organic matter	95	5.0	88	2.5	NS
High EC Control @ 18.0 mS/cm	92	2.8	97	2.8	NS
Carquinez Strait, West of Benicia army dock (405) ³	92	4.8	95	5.0	NS
San Joaquin River between Hog and Turner Cuts (910)	100	0.0	100	0.0	NS
San Joaquin River West of Oulton Pt. (812)	100	0.0	92	5.3	NS
Field Duplicate: Sacramento R. Deep Water Channel, Light 55	100	0.0	-	-	NA
Old River at Holland Cut (915)	98	2.5	100	0.0	NS
Sacramento River Deep Water Channel, Light 55	90	10.0	90	4.1	NS
Sacramento River at Grand Island (711)	79	6.1	75	9.6	NS
Old River, Western arm at railroad bridge (902)	95	2.9	95	3.1	NS

	MSD	PMSD
One-way ANOVA	21.66	22.80
Two-way ANOVA	24.09	25.35

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.082	0.002	-	-	NA
DIEPAMHR + organic matter	0.085	0.009	0.057	0.009	NS
High EC Control @ 18.0 mS/cm	0.053	0.009	0.025	0.001	NS
Carquinez Strait, West of Benicia army dock (405) ³	0.064	0.003	0.045	0.005	NS
San Joaquin River between Hog and Turner Cuts (910)	0.111	0.003	0.063	0.006	S (57%)
San Joaquin River West of Oulton Pt. (812)	0.108	0.006	0.083	0.009	NS
Field Duplicate: Sacramento R. Deep Water Channel, Light 55	0.100	0.007	-	-	NA
Old River at Holland Cut (915)	0.106	0.002	0.090	0.004	NS
Sacramento River Deep Water Channel, Light 55	0.097	0.012	0.083	0.006	NS
Sacramento River at Grand Island (711)	0.088	0.008	0.075	0.004	NS
Old River, Western arm at railroad bridge (902)	0.087	0.003	0.089	0.008	NS

	MSD	PMSD
One-way ANOVA	0.032	38.67
Two-way ANOVA	0.030	40.56

- Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$). Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
- NS: Nonsignificant, S: Significant, NA: Not applicable.
- This high conductivity sample was compared to the High EC control.

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Table A55-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 1/03/07 - 1/04/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Carquinez Strait, West of Benicia army dock (405)	17350	8.6	7.46	11.8	10.3	0.140	0.000
San Joaquin River between Hog and Turner Cuts (910)	498	9.0	7.53	11.8	9.3	0.250	0.001
San Joaquin River West of Oulton Pt. (812)	705	8.8	7.66	12.2	12.0	0.130	0.001
Field Duplicate: Sacramento R. Deep Water Channel, Light 55	355	9.3	7.81	12.3	28.8	0.190	0.002
Old River at Holland Cut (915)	721	8.7	7.61	12.2	5.5	0.900	0.006
Sacramento River Deep Water Channel, Light 55	490	8.6	7.85	12.2	28.4	0.200	0.002
Sacramento River at Grand Island (711)	179	8.4	7.46	12.1	14.0	0.360	0.002
Old River, Western arm at railroad bridge (902)	820	8.9	7.51	12.0	4.7	0.070	0.000

Table A55-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 1/05/2007 of samples collected by the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWS) on 1/03/2007-1/04/2007.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	397	22.1	22.7	7.1	8.9	7.92	8.23	92	60	-
DIEPAMHR + nutrient addback	383	21.1	22.6	7.2	8.8	7.85	8.20	-	-	-
High EC Control @ 18.0 mS	19065	21.3	22.3	7.1	8.6	7.58	7.87	204	10	-
Carquinez Strait, West of Benicia army dock (405)	19115	21.5	22.5	6.8	8.9	7.59	7.87	1960	120	0.002
San Joaquin River between Hog and Turner Cuts (910)	519	21.4	22.4	7.1	8.9	7.93	8.10	112	84	0.010
San Joaquin River West of Oulton Pt. (812)	758	21.6	22.4	7.0	8.8	7.78	8.21	112	72	0.008
Field Duplicate: Sacramento R. Deep Water Channel, Light 55	274	22.1	22.4	7.1	8.7	7.98	8.14	88	90	0.011
Old River at Holland Cut (915)	769	22.0	22.6	7.2	8.9	7.85	8.01	112	70	0.036
Sacramento River Deep Water Channel, Light 55	392	21.6	22.6	7.2	8.7	7.96	8.13	96	90	0.010
Sacramento River at Grand Island (711)	232	21.7	22.7	7.0	8.9	7.79	8.10	72	70	0.018
Old River, Western arm at railroad bridge (902)	831	21.2	22.6	7.1	8.8	7.82	8.11	124	72	0.003
DIEPAMHR + nutrient addback + 25 ppb PBO	403	22.1	22.6	7.4	8.7	7.85	8.04	-	-	-
High EC Control @ 18.0 mS + 25 ppb PBO	19000	22.0	22.7	7.1	8.6	7.69	7.83	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	18515	22.3	22.7	6.9	8.7	7.71	7.86	-	-	-
San Joaquin River between Hog and Turner Cuts (910) + 25 ppb PBO	540	22.2	22.6	7.3	8.7	8.05	8.12	-	-	-
San Joaquin River West of Oulton Pt. (812) + 25 ppb PBO	749	22.1	22.4	7.2	8.7	7.98	8.06	-	-	-
Old River at Holland Cut (915) + 25 ppb PBO	776	22.5	22.5	7.1	8.9	7.89	8.03	-	-	-
Sacramento River Deep Water Channel, Light 55 + 25 ppb PBO	288	22.4	22.8	7.1	8.9	7.92	8.11	-	-	-
Sacramento River at Grand Island (711) + 25 ppb PBO	206	22.2	22.7	7.0	8.6	7.86	8.06	-	-	-
Old River, Western arm at railroad bridge (902) + 25 ppb PBO	878	22.1	22.5	7.3	8.7	7.88	8.01	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A56-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 01/18/2007 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 1/16/07 - 1/17/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	100	0.0	-	-	NA
DIEPAMHR + organic matter	98	2.5	92	2.6	NS
High EC Control @ 11.5 mS/cm	100	0.0	100	0.0	NS
High EC Control @ 16.5 mS/cm	88	7.5	95	3.1	NS
High EC Control @ 24.0 mS/cm	74	6.1	81	9.6	NS
Napa River @ Riverside Blvd. Term. (340)	100	0.0	100	0.0	NS
Suisun Bay, off Chipps Island (508)	100	0.0	95	2.9	NS
Suisun Bay, East of middle point (504) ³	98	2.5	90	7.1	NS
Carquinez Strait, West of Benicia army dock (405) ⁵	92	8.3	76	7.7	NS
Grizzly Bay @ Dolphin (602) ⁴	100	0.0	95	2.9	NS
Montezuma Slough @ Nurse Slough (609) ³	100	0.0	98	2.5	NS
Trip Blank (827)	98	2.5	-	-	NA

	MSD	PMSD
One-way ANOVA	19.26	19.76
Two-way ANOVA	23.87	24.48

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.063	0.004	-	-	NA
DIEPAMHR + organic matter	0.076	0.005	0.045	0.011	NS
High EC Control @ 11.5 mS/cm	0.052	0.005	0.046	0.004	NS
High EC Control @ 16.5 mS/cm	0.045	0.003	0.050	0.003	NS
High EC Control @ 24.0 mS/cm	0.051	0.002	0.033	0.005	NS
Napa River @ Riverside Blvd. Term. (340)	0.099	0.005	0.079	0.015	NS
Suisun Bay, off Chipps Island (508)	0.091	0.004	0.065	0.005	NS
Suisun Bay, East of middle point (504) ³	0.088	0.008	0.040	0.009	S (45%)
Carquinez Strait, West of Benicia army dock (405) ⁵	0.049	0.006	0.035	0.003	NS
Grizzly Bay @ Dolphin (602) ⁴	0.072	0.003	0.059	0.007	NS
Montezuma Slough @ Nurse Slough (609) ³	0.084	0.005	0.054	0.002	NS
Trip Blank (827)	0.066	0.002	-	-	NA

	MSD	PMSD
One-way ANOVA	0.023	30.72
Two-way ANOVA	0.033	43.61

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$). Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
2. NS: Nonsignificant, S: Significant, NA: Not applicable.
3. These high conductivity samples were compared to the High EC control @ 11.5 mS/cm.
4. This high conductivity sample was compared to the High EC control @ 16.5 mS/cm.
5. This high conductivity sample was compared to the High EC control @ 24.0 mS/cm.

Table A56-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 1/16/07 - 1/17/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total	Unionized
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)		Ammonia Nitrogen (mg/L)	Ammonia (mg/L)
Napa River @ Riverside Blvd. Term. (340)	8280	6.5	7.74	11.8	8.0	0.33	0.002
Suisun Bay, off Chipps Island (508)	5540	7.2	7.85	13.0	11.9	0.19	0.002
Suisun Bay, East of middle point (504)	10650	7.2	8.07	12.5	12.8	0.2	0.003
Carquinez Strait, West of Benicia army dock (405)	23560	7.9	8.15	12.0	26.9	0.17	0.003
Grizzly Bay @ Dolphin (602)	16320	7.4	8.08	12.5	22.0	0.21	0.003
Montezuma Slough @ Nurse Slough (609)	11300	6.7	7.82	12.4	29.2	0.26	0.002
Trip Blank (827)	338	13.4	7.91	11.4	0.2	0	0.000

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Table A56-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 1/18/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 1/16/07- 1/17/07.

Treatment	Laboratory Chemistry						Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹	
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH				Max pH
DIEPAMHR	359	22.4	23.5	7.3	8.8	7.92	8.01	106	56	-
DIEPAMHR + organic matter	362	22.5	24.1	7.3	8.6	7.79	8.10	106	56	-
High EC Control @ 11.5 mS/cm	11450	22.5	23.7	7.3	8.8	7.55	7.93	1280	70	-
High EC Control @ 16.5 mS/cm	16655	22.5	24.3	7.3	8.7	7.63	7.90	1890	78	-
High EC Control @ 24.0 mS/cm	23890	22.4	24.2	7.0	8.8	7.65	7.97	2780	89	-
Napa River @ Riverside Blvd. Term. (340)	8345	22.4	24.3	7.5	8.8	7.86	7.91	940	113	0.009
Suisun Bay, off Chipps Island (508)	5660	22.3	24.4	7.6	8.9	7.82	7.97	630	83	0.007
Suisun Bay, East of middle point (504)	10680	22.3	24.3	7.2	8.9	7.68	7.73	1170	87	0.004
Carquinez Strait, West of Benicia army dock (405)	23380	22.3	23.0	7.1	8.8	7.62	7.71	2720	99	0.003
Grizzly Bay @ Dolphin (602)	16690	22.0	23.8	7.4	8.7	7.71	7.79	860	93	0.004
Montezuma Slough @ Nurse Slough (609)	11400	22.6	23.5	7.5	8.6	7.70	7.78	1290	97	0.005
Trip Blank (827)	570	22.5	23.8	7.5	8.8	7.84	8.18	104	59	0.000
DIEPAMHR + organic matter + 25 ppb BBO	359	21.7	24.2	7.3	8.6	7.79	8.07	-	-	-
High EC Control @ 11.5 mS/cm + 25 ppb PBO	11175	22.3	23.4	7.5	8.9	7.57	7.77	-	-	-
High EC Control @ 16.5 mS/cm + 25 ppb PBO	16035	22.2	23.7	7.3	8.8	7.58	7.84	-	-	-
High EC Control @ 24.0 mS/cm + 25 ppb PBO	23130	22.1	23.9	7.2	8.6	7.61	7.88	-	-	-
Napa River @ Riverside Blvd. Term. (340) + 25 ppb PBO	8620	22.0	24.3	7.0	8.7	7.85	8.04	-	-	-
Suisun Bay, off Chipps Island (508) + 25 ppb PBO	5645	22.0	24.2	7.1	8.9	7.83	8.05	-	-	-
Suisun Bay, East of middle point (504) + 25 ppb PBO	10735	21.9	24.1	7.3	8.6	7.69	7.75	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	22930	21.8	24.1	7.4	8.4	7.64	7.74	-	-	-
Grizzly Bay @ Dolphin (602) + 25 ppb PBO	16500	21.9	24.2	7.2	8.5	7.73	7.84	-	-	-
Montezuma Slough @ Nurse Slough (609) + 25 ppb PBO	11460	21.7	23.9	7.3	8.9	7.79	7.85	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A57-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 01/19/2007 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 1/18/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	95	2.9	-	-	NA
DIEPAMHR + organic matter	90	4.1	95	2.9	NS
Sacramento River @ Grand Island (711)	100	0.0	94	3.4	NS
Old River, Western arm at railroad bridge (902)	100	0.0	98	2.3	NS
San Joaquin River between Hog and Turner Cut (910)	100	0.0	100	0.0	NS
Broad Slough, West end (804)	100	0.0	100	0.0	NS
San Joaquin River, West of Oulton Point (812)	100	0.0	100	0.0	NS
Old River @ Holland Cut (915)	98	2.5	100	0.0	NS
Sacramento River, across from Sherman Lake (704)	100	0.0	98	2.5	NS
Sacramento River Deep Water Channel, Light 55	100	0.0	100	0.0	NS

	MSD	PMSD
One-way ANOVA	8.82	9.80
Two-way ANOVA	9.13	9.87

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.042	0.009	-	-	NA
DIEPAMHR + organic matter	0.071	0.005	0.028	0.003	S (39%)
Sacramento River @ Grand Island (711)	0.043	0.005	0.031	0.010	NS
Old River, Western arm at railroad bridge (902)	0.079	0.001	0.048	0.007	NS
San Joaquin River between Hog and Turner Cut (910)	0.083	0.009	0.050	0.004	NS
Broad Slough, West end (804)	0.091	0.007	0.054	0.005	NS
San Joaquin River, West of Oulton Point (812)	0.077	0.008	0.042	0.010	NS
Old River @ Holland Cut (915)	0.084	0.011	0.057	0.012	NS
Sacramento River, across from Sherman Lake (704)	0.102	0.008	0.054	0.011	S (53%)
Sacramento River Deep Water Channel, Light 55	0.082	0.007	0.056	0.013	NS

	MSD	PMSD
One-way ANOVA	0.036	50.36
Two-way ANOVA	0.041	82.85

- Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$). Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
- NS: Nonsignificant, S: Significant, NA: Not applicable

Table A57-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 1/18/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Un-ionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Sacramento River @ Grand Island (711)	190	6.1	8.00	13.4	9.1	0.18	0.002
Old River, Western arm at railroad bridge (902)	520	5.8	7.95	12.9	7.4	0.11	0.001
San Joaquin River between Hog and Turner Cut (910)	590	6.6	7.91	12.9	6.5	0.21	0.002
Broad Slough, West end (804)	2976	7.2	7.70	12.5	16.6	0.21	0.001
San Joaquin River, West of Oulton Point (812)	464	6.7	8.00	13.1	8.9	0.13	0.002
Old River @ Holland Cut (915)	522	6.6	7.91	12.9	8.5	0.09	0.001
Sacramento River, across from Sherman Lake (704)	1983	7.2	7.80	12.4	18.9	0.21	0.002
Sacramento River Deep Water Channel, Light 55	281	6.4	8.30	13.2	22.5	0.18	0.005

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Table A57-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 1/19/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 1/18/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	370	22.1	24.2	7.5	8.7	7.71	8.01	106	56	-
DIEPAMHR + organic matter	375	22.3	23.5	7.1	8.6	7.63	7.86	106	56	-
Sacramento River @ Grand Island (711)	219	22.5	24.6	7.3	8.8	7.83	7.88	72	74	0.006
Old River, Western arm at railroad bridge (902)	547	22.5	24.3	7.4	8.7	7.78	7.84	100	76	0.004
San Joaquin River between Hog and Turner Cut (910)	636	22.4	24.8	7.2	8.8	7.75	7.83	128	92	0.007
Broad Slough, West end (804)	3085	22.5	24.9	7.5	8.9	7.70	7.74	344	81	0.005
San Joaquin River, West of Oulton Point (812)	600	22.6	24.6	7.2	8.5	7.78	7.99	96	75	0.006
Old River @ Holland Cut (915)	555	22.6	24.8	7.2	8.6	7.77	7.95	102	73	0.004
Sacramento River, across from Sherman Lake (704)	2036	22.5	24.4	7.3	8.9	7.68	7.73	246	82	0.005
Sacramento River Deep Water Channel, Light 55	360	22.4	24.7	7.3	8.8	7.81	7.90	94	100	0.006
DIEPAMHR + organic matter + 25 ppb PBO	384	22.4	23.5	7.3	8.7	7.68	7.76	-	-	-
Sacramento River @ Grand Island (711) + 25 ppb PBO	217	22.5	24.1	7.3	8.9	7.76	7.83	-	-	-
Old River, Western arm at railroad bridge (902) + 25 ppb PBO	550	22.5	24.2	7.1	8.5	7.72	7.87	-	-	-
San Joaquin River between Hog and Turner Cut (910) + 25 ppb PBO	634	22.5	24.2	7.4	8.7	7.69	7.85	-	-	-
Broad Slough, West end (804) + 25 ppb PBO	2948	22.5	23.5	7.5	8.7	7.62	7.79	-	-	-
San Joaquin River, West of Oulton Point (812) + 25 ppb PBO	552	22.4	23.5	7.3	8.7	7.78	8.06	-	-	-
Old River @ Holland Cut (915) + 25 ppb PBO	551	22.5	23.4	7.3	8.8	7.75	7.97	-	-	-
Sacramento River, across from Sherman Lake (704) + 25 ppb PBO	2053	22.3	24.2	7.4	8.7	7.70	7.83	-	-	-
Sacramento River Deep Water Channel, Light 55 + 25 ppb PBO	360	22.3	24.7	6.9	8.9	7.49	7.93	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A58-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 2/02/2007 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 1/31/07 - 2/01/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO ²
DIEPAMHR	100	0.0	-	-	NA
DIEPAMHR + organic matter	100	0.0	83	14.4	NS
High EC Control @ 15.0 mS/cm + organic matter	100	0.0	88	2.5	NS
Grizzly Bay @ Dolphin (602) ³	100	0.0	100	0.0	NS
Old River, Western arm at railroad bridge (902)	100	0.0	100	0.0	NS
Old River @ Holland Cut (915)	100	0.0	98	2.5	NS
San Joaquin River between Hog and Turner Cut (910)	100	0.0	98	2.3	NS
Sacramento River @ Grand Island (711)	90	7.1	77	9.3	NS
Sacramento River Deep Water Channel, Light 55	77	9.4	95	2.8	NS
Sacramento River across from Sherman Lake (704)	100	0.0	93	4.4	NS
San Joaquin River, West of Oulton Point (812)	100	0.0	98	2.3	NS

	MSD	PMSD
One-way ANOVA	17.48	17.48
Two-way ANOVA	25.74	25.74

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO ²
DIEPAMHR	0.083	0.005	-	-	NA
DIEPAMHR + organic matter	0.107	0.009	0.090	0.010	NS
High EC Control @ 15.0 mS/cm + organic matter	0.097	0.007	0.069	0.009	NS
Grizzly Bay @ Dolphin (602) ³	0.100	0.005	0.071	0.004	NS
Old River, Western arm at railroad bridge (902)	0.130	0.010	0.090	0.003	NS
Old River @ Holland Cut (915)	0.137	0.009	0.092	0.007	S (68%)
San Joaquin River between Hog and Turner Cut (910)	0.137	0.002	0.097	0.005	S (71%)
Sacramento River @ Grand Island (711)	0.118	0.005	0.103	0.009	NS
Sacramento River Deep Water Channel, Light 55	0.128	0.016	0.090	0.005	NS
Sacramento River across from Sherman Lake (704)	0.092	0.006	0.093	0.003	NS
San Joaquin River, West of Oulton Point (812)	0.086	0.008	0.078	0.004	NS

	MSD	PMSD
One-way ANOVA	0.04	38.72
Two-way ANOVA	0.04	37.22

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$). Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
2. NS: Nonsignificant, S: Significant, NA: Not applicable
3. This high conductivity sample was compared to the High EC control.

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Table A58-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 1/31/07 - 2/01/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Grizzly Bay @ Dolphin (602)	14820	8.9	7.73	11.8	10.3	0.23	0.002
Old River, Western arm at railroad bridge (902)	436	8.1	7.72	12.4	5.3	0.12	0.001
Old River @ Holland Cut (915)	450	8.1	7.68	12.2	5.2	0.13	0.001
San Joaquin River between Hog and Turner Cut (910)	649	8.3	7.80	12.9	4.7	0.19	0.002
Sacramento River @ Grand Island (711)	195	9.2	7.76	12.4	6.5	0.36	0.003
Sacramento River Deep Water Channel, Light 55	295	8.8	7.91	12.3	20.0	0.27	0.003
Sacramento River across from Sherman Lake (704)	2128	8.6	7.79	12.2	18.5	0.3	0.003
San Joaquin River, West of Oulton Point (812)	421	8.2	7.75	12.5	7.5	0.2	0.002

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Table A58-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 2/02/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 1/31/07 - 2/01/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	354	22.4	24.0	6.7	8.6	7.59	8.05	108	59	-
DIEPAMHR +organic matter	346	22.1	23.0	6.5	8.4	7.58	8.01	108	59	-
High EC Control @ 15.0 mS + organic matter	14260	22.4	24.3	6.8	8.6	7.43	7.74	336	74	-
Grizzly Bay @ Dolphin (602)	14645	22.6	24.6	6.9	8.5	7.55	7.72	332	95	0.004
Old River, Western arm at railroad bridge (902)	497	22.1	23.6	6.6	8.5	7.67	8.00	108	74	0.005
Old River @ Holland Cut (915)	448	22.0	24.6	6.8	8.7	7.60	7.85	100	74	0.004
San Joaquin River between Hog and Turner Cut (910)	601	22.4	24.7	6.6	8.7	7.67	7.88	140	86	0.007
Sacramento River @ Grand Island (711)	199	22.2	24.6	6.8	8.5	7.61	7.95	64	70	0.016
Sacramento River Deep Water Channel, Light 55	283	22.2	23.9	6.5	8.6	7.70	7.97	104	96	0.012
Sacramento River across from Sherman Lake (704)	1991	22.5	23.6	7.0	8.6	7.60	7.89	252	80	0.010
San Joaquin River, West of Oulton Point (812)	399	22.2	24.5	6.7	8.5	7.63	7.97	92	76	0.009
DIEPAMHR + organic matter + 25 ppb PBO	365	22.5	23.2	6.7	8.4	7.53	8.03	-	-	-
High EC Control @ 15.0 mS + organic matter + 25 ppb PBO	14155	22.0	24.0	6.6	8.3	7.44	7.84	-	-	-
Grizzly Bay @ Dolphin (602) + 25 ppb PBO	14225	22.5	23.7	6.7	8.3	7.51	7.75	-	-	-
Old River, Western arm at railroad bridge (902) + 25 ppb PBO	481	22.1	24.2	6.3	8.4	7.62	8.17	-	-	-
Old River @ Holland Cut (915) + 25 ppb PBO	444	22.5	23.8	6.6	8.6	7.67	7.90	-	-	-
San Joaquin River between Hog and Turner Cut (910) + 25 ppb PBO	620	22.1	23.9	6.6	8.6	7.67	7.92	-	-	-
Sacramento River @ Grand Island (711) + 25 ppb PBO	197	22.4	23.9	6.5	8.7	7.66	7.98	-	-	-
Sacramento River Deep Water Channel, Light 55 + 25 ppb PBO	292	22.4	24.6	6.6	8.4	7.69	7.91	-	-	-
Sacramento River across from Sherman Lake (704) + 25 ppb PBO	2087	22.0	23.3	6.8	8.5	7.56	7.86	-	-	-
San Joaquin River, West of Oulton Point (812) + 25 ppb PBO	414	21.8	24.3	6.8	8.4	7.59	7.99	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A59-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 2/07/07 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 1/30/07 - 1/31/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ³
	x	se	x	se	
DIEPAMHR	100	0.0	-	-	NA
DIEPAMHR + organic matter	95	2.9	89	4.5	NS
High EC Control @ 12.0 mS/cm + organic matter	97	2.8	98	2.5	NS
High EC Control @ 24.0 mS/cm + organic matter	88	6.3	63	7.6	S (72%)
Napa River @ Riverside Blvd Terminus (340) ³	100	0.0	100	0.0	NS
Suisun Bay, East of middle point (504) ³	100	0.0	-	-	NA
Montezuma Slough @ Nurse Slough (609)	100	0.0	-	-	NA
Broad Slough, West end (804)	100	0.0	-	-	NA
Carquinez Strait, West of Benicia army ferry dock (405) ⁴	92	2.8	-	-	NA
Suisun Bay, off Chipps Island (508)	98	2.5	-	-	NA
Field Duplicate: Napa River @ Riverside Blvd Terminus (340) ³	97	2.8	-	-	NA
Mixed Water: 504 + 508	-	-	100	0.0	NA
Mixed Water: 609 + 504	-	-	100	0.0	NA
Mixed Water: 804 + 405	-	-	100	0.0	NA
Mixed Water: 405 + 804	-	-	84	3.5	NA
Mixed Water: 508 + 609	-	-	87	4.9	NA

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ³
	x	se	x	se	
DIEPAMHR	0.053	0.003	-	-	NA
DIEPAMHR + organic matter	0.087	0.007	0.057	0.003	NS
High EC Control @ 12.0 mS/cm + organic matter	0.080	0.005	0.043	0.008	NS
High EC Control @ 24.0 mS/cm + organic matter	0.023	0.009	0.006	0.022	NS
Napa River @ Riverside Blvd Terminus (340) ³	0.061	0.012	0.072	0.005	NS
Suisun Bay, East of middle point (504) ³	0.063	0.008	-	-	NA
Montezuma Slough @ Nurse Slough (609)	0.081	0.004	-	-	NA
Broad Slough, West end (804)	0.075	0.006	-	-	NA
Carquinez Strait, West of Benicia army ferry dock (405) ⁴	0.049	0.007	-	-	NA
Suisun Bay, off Chipps Island (508)	0.078	0.007	-	-	NA
Field Duplicate: Napa River @ Riverside Blvd Terminus (340) ³	0.081	0.007	-	-	NA
Mixed Water: 504 + 508	-	-	0.057	0.012	NA
Mixed Water: 609 + 504	-	-	0.084	0.006	NA
Mixed Water: 804 + 405	-	-	0.088	0.003	NA
Mixed Water: 405 + 804	-	-	0.044	0.011	NA
Mixed Water: 508 + 609	-	-	0.080	0.012	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$). Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
2. NS: Nonsignificant, S: Significant, NA: Not applicable.
3. These high conductivity samples were compared to the 12.0 mS/cm High EC control.
4. This high conductivity sample was compared to the 24.0 mS/cm High EC control.

Table A59-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 1/30/07 - 1/31/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Napa River @ Riverside Blvd Terminus (340)	12080	8.0	8.05	13.5	12.9	0.04	0.001
Suisun Bay, East of middle point (504)	10960	8.4	7.7	12.2	9.5	0.22	0.000
Montezuma Slough @ Nurse Slough (609)	5020	8.5	7.58	11.9	31.7	0.20	0.001
Broad Slough, West end (804)	1376	8.2	7.67	12.9	4.4	0.20	0.001
Carquinez Strait, West of Benicia army ferry dock (405)	22830	9.2	7.72	11.2	16.1	0.20	0.001
Suisun Bay, off Chipps Island (508)	4775	8.2	7.71	12.7	11.8	0.20	0.001
Field Duplicate: Napa River @ Riverside Blvd Terminus (340)	-	-	-	-	9.4	0.05	-

Table A59-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 2/07/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 1/30/07 - 1/31/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	339	22.2	24.3	7.6	8.4	7.61	7.94	104	64	-
DIEPAMHR + 1% nutrient addback	339	22.5	24.4	7.2	8.3	7.44	8.04	104	64	-
High EC Control @ 12.0 mS/cm + nutrient addback	11285	22.5	24.5	7.6	8.4	7.49	7.65	1350	70	-
High EC Control @ 24.0 mS/cm + nutrient addback	22915	22.5	24.5	7.7	8.6	7.55	7.66	2740	86	-
Napa River @ Riverside Blvd Terminus (340)	11915	22.5	24.8	7.3	8.6	7.68	7.82	1340	116	0.001
Suisun Bay, East of middle point (504)	10935	22.5	24.7	7.2	8.7	7.60	7.81	1230	90	0.004
Montezuma Slough @ Nurse Slough (609)	4955	22.4	24.4	7.3	8.7	7.66	7.90	560	85	0.005
Broad Slough, West end (804)	1434	22.5	24.8	7.4	8.6	7.69	8.04	188	80	0.008
Carquinez Strait, West of Benicia army ferry dock (405)	23455	22.7	24.5	7.4	8.9	7.45	7.68	2740	103	0.002
Suisun Bay, off Chipps Island (508)	4766	22.9	24.4	7.4	8.3	7.67	7.99	560	87	0.007
Field Duplicate: Napa River @ Riverside Blvd Terminus (340)	11730	22.5	24.6	7.6	8.6	7.74	7.81	1380	115	0.001
DIEPAMHR + 1% nutrient addback + 25 ppb PBO	364	22.8	24.1	7.2	8.7	7.65	8.15	NA	NA	-
High EC Control @ 12.0 mS/cm + nutrient addback + 25 ppb PBO	11500	22.9	24.0	7.7	8.8	7.56	7.67	NA	NA	-
High EC Control @ 24.0 mS/cm + nutrient addback + 25 ppb PBO	22730	22.7	23.9	7.6	8.4	7.53	7.74	NA	NA	-
Napa River @ Riverside Blvd Terminus (340) + 25 ppb PBO	11640	23.2	24.0	7.5	8.9	7.76	7.89	NA	NA	-
Mixed Water (504 + 508) + 25 ppb PBO	8410	23.2	23.6	7.3	8.9	7.60	7.82	NA	NA	-
Mixed Water (609 + 504) + 25 ppb PBO	7219	23.1	23.3	7.1	8.8	7.55	7.92	NA	NA	-
Mixed Water (804 + 405) + 25 ppb PBO	9763	23.0	23.2	7.6	8.1	7.61	8.14	NA	NA	-
Mixed Water (405 + 804) + 25 ppb PBO	15360	23.0	23.2	7.6	8.7	7.57	7.68	NA	NA	-
Mixed Water (508 + 609) + 25 ppb PBO	4723	22.8	23.2	7.4	8.8	7.64	8.01	NA	NA	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A60-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 2/15/2007 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 2/13/07 - 2/14/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	98	2.5	-	-	NA
DIEPAMHR + organic matter	93	4.8	100	0.0	NS
High EC Control @ 15.95 mS/cm + organic matter	100	0.0	90	7.1	NS
Napa River @ Riverside Blvd terminus (340)	100	0.0	98	2.5	NS
Suisun Bay, off Chipps Island (508)	95	5.0	98	2.5	NS
Grizzly Bay @ Dolphin (602)	100	0.0	100	0.0	NS
Montezuma Slough @ Nurse Slough (609)	100	0.0	100	0.0	NS
Carquinez Strait, West of Benicia army dock (405) ³	93	4.8	98	2.5	NS
Broad Slough, West End (804)	98	2.5	100	0.0	NS
Suisun Bay, East of middle point (504)	100	0.0	100	0.0	NS
Bottle Blank	98	2.5	-	-	NA

	MSD	PMSD
One-way ANOVA	14.03	15.17
Two-way ANOVA	14.94	16.15

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.059	0.002	-	-	NA
DIEPAMHR + organic matter	0.053	0.004	0.052	0.005	NS
High EC Control @ 15.95 mS/cm + organic matter	0.061	0.003	0.032	0.006	NS
Napa River @ Riverside Blvd terminus (340)	0.098	0.011	0.063	0.006	S (64%)
Suisun Bay, off Chipps Island (508)	0.091	0.004	0.073	0.008	NS
Grizzly Bay @ Dolphin (602)	0.079	0.004	0.065	0.008	NS
Montezuma Slough @ Nurse Slough (609)	0.083	0.006	0.061	0.005	NS
Carquinez Strait, West of Benicia army dock (405) ³	0.066	0.003	0.054	0.005	NS
Broad Slough, West End (804)	0.079	0.007	0.054	0.006	NS
Suisun Bay, East of middle point (504)	0.074	0.003	0.063	0.014	NS
Bottle Blank	0.045	0.001	-	-	NA

	MSD	PMSD
One-way ANOVA	0.03	48.66
Two-way ANOVA	0.03	59.35

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable

3. This high conductivity sample was compared to the High EC control.

Table A60-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 2/13/07 - 2/14/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Napa River @ Riverside Blvd terminus (340)	334	11.1	7.10	10.2	64.3	0.09	0.000
Suisun Bay, off Chipps Island (508)	921	10.7	7.45	10.8	10.7	0.21	0.001
Grizzly Bay @ Dolphin (602)	8290	10.8	7.77	10.9	16.3	0.20	0.002
Montezuma Slough @ Nurse Slough (609)	3130	11.5	7.47	10.0	23.7	0.20	0.001
Carquinez Strait, West of Benicia army dock (405)	15390	11.1	7.78	10.3	13.2	0.19	0.002
Broad Slough, West End (804)	584	10.8	7.68	11.4	5.9	0.13	0.001
Suisun Bay, East of middle point (504)	1761	10.3	7.65	11.2	11.9	0.22	0.002
Bottle Blank	329	21.2	8.19	9.8	0.3	0.00	0.000

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Table A60-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 2/15/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 2/13/07- 2/14/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	354	20.8	24.2	7.5	8.3	7.90	8.12	108	59	-
DIEPAMHR + organic matter	358	21.4	24.9	7.4	8.2	7.74	8.03	108	59	-
High EC Control @ 15.95 mS/cm + organic matter	14265	21.3	24.7	7.7	8.9	7.63	7.77	-	-	-
Napa River @ Riverside Blvd terminus (340)	4890	21.3	24.8	7.4	8.5	7.81	8.17	102	57	0.005
Suisun Bay, off Chipps Island (508)	1274	21.3	24.9	7.5	8.5	7.98	8.07	144	81	0.011
Grizzly Bay @ Dolphin (602)	8245	21.4	24.6	7.7	8.3	7.80	7.86	910	88	0.006
Montezuma Slough @ Nurse Slough (609)	3158	21.0	24.7	7.7	8.3	7.91	7.99	370	84	0.008
Carquinez Strait, West of Benicia army dock (405)	15300	21.4	24.8	7.3	8.2	7.67	7.73	1720	94	0.004
Broad Slough, West End (804)	781	21.3	24.6	7.5	8.3	8.04	8.15	112	80	0.009
Suisun Bay, East of middle point (504)	2441	21.4	24.5	7.7	8.4	7.86	7.98	296	88	0.009
Bottle Blank	422	21.6	24.3	7.6	8.4	7.82	8.07	102	58	0.000
DIEPAMHR + organic matter + 25 ppb PBO	495	20.9	24.4	7.7	8.5	7.93	8.12	-	-	-
High EC Control @ 15.95 mS/cm + organic matter + 25 ppb PBO	15450	21.2	24.0	7.5	8.7	7.52	7.78	-	-	-
Napa River @ Riverside Blvd terminus (340) + 25 ppb PBO	5634	21.2	24.1	7.5	8.3	7.62	8.18	-	-	-
Suisun Bay, off Chipps Island (508) + 25 ppb PBO	1203	21.1	24.2	7.5	8.5	7.88	8.16	-	-	-
Grizzly Bay @ Dolphin (602) + 25 ppb PBO	8165	21.1	24.2	7.5	8.4	7.68	7.84	-	-	-
Montezuma Slough @ Nurse Slough (609) + 25 ppb PBO	3231	21.3	24.2	7.6	8.4	7.88	7.96	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	14830	21.4	24.3	7.5	8.5	7.67	7.70	-	-	-
Broad Slough, West End (804) + 25 ppb PBO	672	21.1	24.2	7.5	8.9	8.04	8.13	-	-	-
Suisun Bay, East of middle point (504) + 25 ppb PBO	2442	20.7	24.1	7.4	8.6	7.83	8.05	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A61-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 2/16/2007 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 2/15/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	92	2.7	-	-	NA
DIEPAMHR + organic matter	78	6.3	100	0.0	S (132%)
San Joaquin River between Hog and Turner Cuts (910)	100	0.0	95	2.9	NS
Sacramento River at tip of Grand Island (711)	89	4.5	86	2.2	NS
San Joaquin River, West of Oulton Point (812)	100	0.0	97	2.8	NS
Sacramento River Deep Water Channel, Light 55	98	2.5	100	0.0	NS
Old River, western arm at railroad bridge (902)	98	2.5	98	2.3	NS
Old River at Holland Cut (915)	100	0.0	100	0.0	NS
Sacramento River, North side across from Sherman Lake (704)	98	2.5	93	4.7	NS

	MSD	PMSD
One-way ANOVA	14.8	19.1
Two-way ANOVA	14.4	18.6

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.048	0.005	-	-	NA
DIEPAMHR + organic matter	0.049	0.007	0.063	0.003	NS
San Joaquin River between Hog and Turner Cuts (910)	0.090	0.006	0.067	0.009	NS
Sacramento River at tip of Grand Island (711)	0.071	0.007	0.042	0.004	NS
San Joaquin River, West of Oulton Point (812)	0.092	0.011	0.061	0.007	NS
Sacramento River Deep Water Channel, Light 55	0.097	0.006	0.040	0.006	S (41%)
Old River, western arm at railroad bridge (902)	0.103	0.006	0.052	0.004	S (50%)
Old River at Holland Cut (915)	0.080	0.001	0.057	0.006	NS
Sacramento River, North side across from Sherman Lake (704)	0.087	0.005	0.069	0.007	NS

	MSD	PMSD
One-way ANOVA	0.031	62.7
Two-way ANOVA	0.033	66.4

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable

Table A61-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 2/15/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
San Joaquin River between Hog and Turner Cuts (910)	702	12.1	7.62	10.5	3.4	0.16	0.001
Sacramento River at tip of Grand Island (711)	121	11.8	7.39	10.1	60.8	0.23	0.001
San Joaquin River, West of Oulton Point (812)	245	11.6	7.51	10.7	9.8	0.16	0.001
Sacramento River Deep Water Channel, Light 55	266	12.4	7.77	10.3	34.8	0.12	0.001
Old River, western arm at railroad bridge (902)	390	11.3	7.49	10.8	3.5	0.05	0.000
Old River at Holland Cut (915)	407	11.5	7.50	10.7	3.7	0.06	0.000
Sacramento River, North side across from Sherman Lake (704)	280	11.9	7.61	10.5	30.3	0.14	0.001

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Table A61-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 2/16/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 2/15/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	336	19.3	25.0	8.1	8.5	7.76	8.04	108	59	-
DIEPAMHR + organic matter	339	19.3	25.0	7.5	8.4	7.68	8.01	108	59	-
San Joaquin River between Hog and Turner Cuts (910)	673	19.4	25.2	7.5	8.6	7.71	7.91	156	96	0.006
Sacramento River at tip of Grand Island (711)	147	19.4	25.0	7.3	8.6	7.65	7.88	52	46	0.009
San Joaquin River, West of Oulton Point (812)	246	19.3	25.2	7.2	8.9	7.61	7.85	76	79	0.006
Sacramento River Deep Water Channel, Light 55	273	19.4	25.4	7.2	8.6	7.67	7.94	92	95	0.005
Old River, western arm at railroad bridge (902)	377	19.4	25.1	7.3	8.7	7.66	7.85	98	78	0.002
Old River at Holland Cut (915)	399	19.4	25.0	7.1	8.4	7.64	7.99	102	79	0.003
Sacramento River, North side across from Sherman Lake (704)	282	19.3	24.8	7.3	8.1	7.65	7.98	74	70	0.007
DIEPAMHR + organic matter + 25 ppb PBO	332	19.4	24.2	8.1	8.4	7.95	8.21	-	-	-
San Joaquin River between Hog and Turner Cuts (910) + 25 ppb PBO	671	19.4	24.6	7.7	8.5	7.80	7.98	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	145	19.4	24.4	7.0	8.6	7.66	7.85	-	-	-
San Joaquin River, West of Oulton Point (812) + 25 ppb PBO	244	19.4	24.4	7.3	8.7	7.62	7.77	-	-	-
Sacramento River Deep Water Channel, Light 55 + 25 ppb PBO	273	19.2	24.5	7.3	8.9	7.74	8.07	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	371	19.2	24.5	7.3	8.9	7.68	7.96	-	-	-
Old River at Holland Cut (915) + 25 ppb PBO	392	19.2	24.6	7.2	8.7	7.67	7.83	-	-	-
Sacramento River, North side across from Sherman Lake (704) + 25 ppb PBO	271	19.2	24.6	7.3	8.9	7.65	7.93	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A62-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 3/01/2007 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 2/28/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO ²
DIEPAMHR	100	0.0	-	-	NS
DIEPAMHR + organic matter	100	0.0	95	2.8	NS
Napa River at Riverside Blvd Terminus (340)	98	2.5	96	3.8	NS
Old River, Western arm at Railroad Bridge (902)	100	0.0	92	4.8	NS
Old River at mouth of Holland Cut (915)	100	0.0	98	2.5	NS
San Joaquin R., between Hog and Turner Cut (910)	100	0.0	100	0.0	NS
San Joaquin R., West of Oulton Point (812)	100	0.0	95	2.9	NS
Sacramento R. at tip of Grand Island (711)	78	6.4	75	4.4	NS
Sacramento R. Deep Water Channel, Light 55	100	0.0	95	3.1	NS
Sacramento R. North side across from Sherman Lake (704)	100	0.0	98	2.1	NS
Broad Slough, West End (804)	98	2.5	98	2.5	NS

	MSD	PMSD
One-way ANOVA	10.9	10.9
Two-way ANOVA	14.5	14.9

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO ²
DIEPAMHR	0.072	0.004	-	-	NS
DIEPAMHR + organic matter	0.090	0.008	0.078	0.005	NS
Napa River at Riverside Blvd Terminus (340)	0.136	0.004	0.102	0.011	NS
Old River, Western arm at Railroad Bridge (902)	0.119	0.014	0.094	0.012	NS
Old River at mouth of Holland Cut (915)	0.116	0.010	0.065	0.005	S (56%)
San Joaquin R., between Hog and Turner Cut (910)	0.090	0.004	0.096	0.009	NS
San Joaquin R., West of Oulton Point (812)	0.110	0.019	0.100	0.009	NS
Sacramento R. at tip of Grand Island (711)	0.065	0.010	0.070	0.007	NS
Sacramento R. Deep Water Channel, Light 55	0.087	0.005	0.081	0.008	NS
Sacramento R. North side across from Sherman Lake (704)	0.074	0.006	0.075	0.006	NS
Broad Slough, West End (804)	0.119	0.013	0.094	0.007	NS

	MSD	PMSD
One-way ANOVA	0.049	54.6
Two-way ANOVA	0.050	58.9

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable

Table A62-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 2/28/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Napa River at Riverside Blvd Terminus (340)	217	9.7	7.55	11.8	36.1	0.06	0.000
Old River, Western arm at Railroad Bridge (902)	293	10.4	7.53	11.3	6.7	0.05	0.000
Old River at mouth of Holland Cut (915)	340	10.9	7.17	11.3	5.8	0.10	0.000
San Joaquin R., between Hog and Turner Cut (910)	653	12.0	7.61	9.9	6.9	0.15	0.001
San Joaquin R., West of Oulton Point (812)	242	11.0	7.63	11.3	9.0	0.12	0.001
Sacramento R. at tip of Grand Island (711)	157	9.6	7.49	11.3	27.0	0.14	0.001
Sacramento R. Deep Water Channel, Light 55	336	10.8	7.92	11.5	20.0	0.12	0.002
Sacramento R. North side across from Sherman Lake (704)	192	10.1	7.63	11.9	11.1	0.19	0.001
Broad Slough, West End (804)	296	10.7	7.66	11.6	10.0	0.10	0.001

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Table A62-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 3/01/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 2/28/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	361	23.2	24.0	7.4	8.7	7.77	8.13	108	60	-
DIEPAMHR +organic matter	366	23.2	24.2	7.2	8.5	7.64	8.06	108	60	-
Napa River at Riverside Blvd Terminus (340)	221	23.3	23.4	7.0	8.9	7.53	8.00	80	59	0.003
Old River, Western arm at Railroad Bridge (902)	295	23.1	24.0	7.1	8.8	7.72	7.99	84	73	0.002
Old River at mouth of Holland Cut (915)	327	23.0	23.6	6.9	8.9	7.70	8.01	94	75	0.005
San Joaquin R., between Hog and Turner Cut (910)	661	23.2	23.8	7.0	8.8	7.63	7.98	152	100	0.006
San Joaquin R., West of Oulton Point (812)	238	23.2	24.2	7.1	8.7	7.66	8.04	80	68	0.006
Sacramento R. at tip of Grand Island (711)	136	23.1	24.2	7.1	8.6	7.61	8.14	66	56	0.009
Sacramento R. Deep Water Channel, Light 55	342	23.1	23.8	7.1	8.6	7.93	8.21	114	104	0.009
Sacramento R. North side across from Sherman Lake (704)	188	23.1	23.4	7.1	8.7	7.61	8.18	72	73	0.013
Broad Slough, West End (804)	294	23.2	23.7	7.3	8.8	7.73	8.10	90	72	0.006
DIEPAMHR + 1% nutrient addback + 25 ppb PBO	360	23.0	23.8	7.3	8.5	7.66	8.15	-	-	-
Napa River at Riverside Blvd Terminus (340) + 25 ppb PBO	223	23.1	24.5	6.9	8.5	7.62	8.09	-	-	-
Old River, Western arm at Railroad Bridge (902) + 25 ppb PBO	285	23.1	24.5	7.0	8.7	7.65	8.04	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	336	22.9	24.5	7.0	8.5	7.73	8.11	-	-	-
San Joaquin R., between Hog and Turner Cut (910) + 25 ppb PBO	653	23.2	24.1	7.1	8.6	7.73	8.03	-	-	-
San Joaquin R., West of Oulton Point (812) + 25 ppb PBO	229	23.4	23.5	7.1	8.6	7.69	8.14	-	-	-
Sacramento R. at tip of Grand Island (711) + 25 ppb PBO	134	23.4	23.9	6.9	8.7	7.53	8.04	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	333	23.1	23.5	7.0	8.7	7.78	8.19	-	-	-
Sacramento R. North side across from Sherman Lake (704) + 25 ppb PBO	187	23.6	24.4	7.0	8.6	7.64	8.06	-	-	-
Broad Slough, West End (804) + 25 ppb PBO	291	24.2	24.2	7.1	8.7	7.71	8.04	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A63-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 3/02/2007 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 3/01/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	98	2.5	-	-	NS
DIEPAMHR + organic matter	98	2.5	98	2.5	NS
Suisun Bay off Chipps Island (508)	100	0.0	90	7.1	NS
Suisun Bay, East of middle point (504)	100	0.0	100	0.0	NS
Carquinez Strait, West of Benicia army dock (405)	95	5.0	100	0.0	NS
Grizzly Bay at Dolphin (602)	95	2.9	98	2.5	NS
Montezuma Slough at Nurse Slough (609)	100	0.0	97	2.8	NS

	MSD	PMSD
One-way ANOVA	11.8	12.1
Two-way ANOVA	15.0	15.4

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.053	0.002	-	-	NS
DIEPAMHR + organic matter	0.084	0.005	0.059	0.011	NS
Suisun Bay off Chipps Island (508)	0.101	0.011	0.061	0.006	S (60%)
Suisun Bay, East of middle point (504)	0.088	0.008	0.068	0.000	NS
Carquinez Strait, West of Benicia army dock (405)	0.107	0.006	0.078	0.005	NS
Grizzly Bay at Dolphin (602)	0.085	0.002	0.091	0.002	NS
Montezuma Slough at Nurse Slough (609)	0.077	0.008	0.078	0.004	NS

	MSD	PMSD
One-way ANOVA	0.032	38.2
Two-way ANOVA	0.033	39.0

- Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.
Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
- NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable

Table A63-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 3/01/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Suisun Bay off Chipps Island (508)	299	10.6	7.81	11.7	16.0	0.2	0.002
Suisun Bay, East of middle point (504)	484	10.9	7.80	11.0	17.8	0.1	0.002
Carquinez Strait, West of Benicia army dock (405)	5100	10.8	7.81	11.3	25.3	0.2	0.002
Grizzly Bay at Dolphin (602)	1196	10.6	7.81	11.5	34.1	0.2	0.002
Montezuma Slough at Nurse Slough (609)	668	10.2	7.57	11.0	13.2	0.3	0.002

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Table A63-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 3/02/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 3/01/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	349	23.3	23.5	7.2	8.8	7.70	8.14	108	60	-
DIEPAMHR + organic matter	352	23.1	23.6	7.0	8.5	7.54	8.20	108	60	-
Suisun Bay off Chipps Island (508)	312	23.5	24.1	7.2	8.8	7.68	8.11	80	78	0.011
Suisun Bay, East of middle point (504)	401	23.0	23.4	7.2	8.8	7.69	8.14	88	76	0.009
Carquinez Strait, West of Benicia army dock (405)	4945	23.5	23.8	7.1	8.5	7.59	7.97	544	80	0.008
Grizzly Bay at Dolphin (602)	1183	23.5	23.8	7.1	8.7	7.70	8.10	164	80	0.011
Montezuma Slough at Nurse Slough (609)	631	23.2	23.4	7.2	8.6	7.66	8.01	120	78	0.013
DIEPAMHR + organic matter + 25 ppb PBO	351	23.4	23.5	7.2	8.5	7.57	8.18	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	317	23.1	23.4	7.1	8.7	7.69	8.16	-	-	-
Suisun Bay, East of middle point (504) + 25 ppb PBO	405	23.5	23.5	7.0	8.9	7.65	8.02	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	4502	23.3	23.4	7.3	8.8	7.60	7.90	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	1185	23.5	23.9	7.2	8.5	7.72	8.06	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	660	23.3	23.8	6.8	8.8	7.57	8.00	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A64-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 3/15/07 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 3/14/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	98	2.5	-	-	NA
DIEPAMHR + organic matter	98	2.5	98	2.5	NS
High EC Control @ 19.0 mS/cm + organic matter	98	2.5	97	2.8	NS
High EC Control @ 11.75 mS/cm + organic matter	98	2.5	95	2.9	NS
Suisun Bay, off Chipps Island (508)	95	2.9	100	0.0	NS
Suisun Bay, East of middle point (504)	98	2.5	98	2.5	NS
Carquinez Strait, West of Benicia army dock (405) ³	95	5.0	90	0.0	NS
Grizzly Bay at Dolphin (602) ⁴	98	2.5	98	2.5	NS
Montezuma Slough at Nurse Slough (609)	98	2.5	100	0.0	NS
Middle of Broad Slough, West end (804)	100	0.0	100	0.0	NS
Napa River at Riverside Blvd terminus (340)	100	0.0	100	0.0	NS

	MSD	PMSD
One-way ANOVA	13.2	13.9
Two-way ANOVA	13.1	13.6

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.078	0.003	-	-	NA
DIEPAMHR + organic matter	0.091	0.007	0.114	0.008	NS
High EC Control @ 19.0 mS/cm + organic matter	0.059	0.004	0.069	0.004	NS
High EC Control @ 11.75 mS/cm + organic matter	0.068	0.006	0.073	0.003	NS
Suisun Bay, off Chipps Island (508)	0.124	0.008	0.123	0.004	NS
Suisun Bay, East of middle point (504)	0.121	0.008	0.109	0.006	NS
Carquinez Strait, West of Benicia army dock (405) ³	0.085	0.003	0.085	0.004	NS
Grizzly Bay at Dolphin (602) ⁴	0.142	0.005	0.106	0.008	S (75%)
Montezuma Slough at Nurse Slough (609)	0.149	0.002	0.114	0.002	S (77%)
Middle of Broad Slough, West end (804)	0.139	0.010	0.124	0.003	NS
Napa River at Riverside Blvd terminus (340)	0.162	0.005	0.152	0.015	NS

	MSD	PMSD
One-way ANOVA	0.030	32.8
Two-way ANOVA	0.034	33.8

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.
Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable
3. This high conductivity sample was compared to the 19.0 mS/cm High EC control.
4. This high conductivity sample was compared to the 11.75 mS/cm High EC control.

Table A64-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 3/14/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Suisun bay, off Chipps Island 508	2665	13.5	7.68	10.4	8.5	0.09	0.001
Suisun Bay, East of middle point (504)	7080	13.4	7.73	10.4	10.2	0.09	0.001
Carquinez Strait, West of Benicia army dock (405)	19180	13.7	7.83	10.1	6.9	0.06	0.001
Grizzly Bay at Dolphin (602)	11790	14.0	7.79	9.8	9.9	0.07	0.001
Montezuma Slough at Nurse Slough (609)	1893	15.2	7.45	8.4	43.4	0.11	0.001
Middle of Broad Slough, West end (804)	267	14.2	7.73	10.2	9.2	0.08	0.001
Napa River at Riverside Blvd terminus (340)	410	17.5	7.97	11.5	11.2	0.00	0.000

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Table A64-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 3/15/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 3/14/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	356	23.1	24.1	7.0	8.7	7.55	7.96	108	62	-
DIEPAMHR + organic matter	362	23.2	24.4	6.8	8.8	7.57	7.92	108	62	-
High EC Control @ 19.0 mS/cm + organic matter	18655	23.3	24.0	7.2	8.6	7.46	7.67	2340	160	-
High EC Control @ 11.75 mS/cm + organic matter	11790	23.3	24.1	7.3	8.9	7.53	7.83	1320	130	-
Suisun Bay, off Chipps Island (508)	2504	22.9	24.3	6.6	8.8	7.60	7.94	296	74	0.003
Suisun Bay, East of middle point (504)	7225	23.2	23.9	6.8	8.6	7.52	7.68	824	76	0.002
Carquinez Strait, West of Benicia army dock (405)	18695	23.2	24.1	6.9	8.8	7.49	7.68	2240	160	0.001
Grizzly Bay at Dolphin (602)	11955	23.2	24.4	7.1	8.7	7.54	8.01	1460	130	0.003
Montezuma Slough at Nurse Slough (609)	1875	22.9	24.3	6.9	8.5	7.67	8.08	256	80	0.003
Middle of Broad Slough, West end (804)	334	23.1	24.3	6.5	8.8	7.56	8.12	72	70	0.001
Napa River at Riverside Blvd terminus (340)	399	23.2	24.4	6.5	8.9	7.83	8.00	112	100	0.000
DIEPAMHR + organic matter + 25 ppb PBO	365	23.1	24.6	6.8	8.3	7.85	7.96	-	-	-
High EC Control @ 19.0 mS/cm + organic matter + 25 ppb PBO	18315	22.8	23.5	7.0	8.1	7.34	7.73	-	-	-
High EC Control @ 11.75 mS/cm + organic matter + 25 ppb PBO	11670	22.8	23.9	7.1	8.6	7.62	7.87	-	-	-
Suisun Bay, off Chipps Island (508) + 25 ppb PBO	2835	22.8	23.9	6.7	8.7	7.60	7.93	-	-	-
Suisun Bay, East of middle point (504) + 25 ppb PBO	7115	23.1	24.2	6.6	8.6	7.42	7.70	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	18650	22.8	23.7	6.7	8.2	7.55	7.77	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	12075	23.3	23.9	6.9	8.5	7.48	7.76	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	1932	22.7	24.4	6.8	8.4	7.79	7.80	-	-	-
Middle of Broad Slough, West end (804) + 25 ppb PBO	298	23.0	24.5	6.8	8.7	7.79	7.94	-	-	-
Napa River at Riverside Blvd terminus (340) + 25 ppb PBO	424	23.5	23.9	6.8	8.7	7.80	7.97	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A65-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 03/17/2007 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 3/16/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	100	0.0	-	-	NA
DIEPAMHR + organic matter	100	0.0	98	2.5	NS
Old River, western arm at railroad bridge (902)	100	0.0	100	0.0	NS
Old River at mouth of Holland Cut (915)	100	0.0	98	2.5	NS
San Joaquin River between Hog and Turner Cut (910)	100	0.0	98	2.3	NS
Sacramento River at tip of Grand Island (711)	100	0.0	98	2.5	NS
Sacramento River Deep Water Channel, Light 55	90	7.1	98	2.5	NS
Sacramento River across from Sherman Lake (704)	100	0.0	100	0.0	NS
San Joaquin River, West of Oulton Point (812)	100	0.0	100	0.0	NS
Field Duplicate: Sacramento River across from Sherman Lake (704)	100	0.0	-	-	NA

	MSD	PMSD
One-way ANOVA	10.8	10.8
Two-way ANOVA	11.4	11.6

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.077	0.003	-	-	NA
DIEPAMHR + organic matter	0.107	0.008	0.068	0.006	NS
Old River, western arm at railroad bridge (902)	0.114	0.010	0.081	0.004	NS
Old River at mouth of Holland Cut (915)	0.116	0.005	0.089	0.011	NS
San Joaquin River between Hog and Turner Cut (910)	0.101	0.010	0.093	0.004	NS
Sacramento River at tip of Grand Island (711)	0.091	0.007	0.064	0.003	NS
Sacramento River Deep Water Channel, Light 55	0.105	0.013	0.094	0.007	NS
Sacramento River across from Sherman Lake (704)	0.096	0.013	0.086	0.004	NS
San Joaquin River, West of Oulton Point (812)	0.108	0.005	0.095	0.010	NS
Field Duplicate: Sacramento River across from Sherman Lake (704)	0.090	0.005	-	-	NA

	MSD	PMSD
One-way ANOVA	0.042	39.1
Two-way ANOVA	0.042	47.6

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$). Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable

Table A65-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 3/16/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Old River, western arm at railroad bridge (902)	233	16.6	7.46	10.4	5.5	0.00	0.000
Old River at mouth of Holland Cut (915)	245	16.7	7.38	10.2	7.9	0.01	0.000
San Joaquin River between Hog and Turner Cut (910)	456	17.5	7.28	9.4	5.1	0.05	0.000
Sacramento River at tip of Grand Island (711)	167	15.9	7.35	10.7	4.9	0.30	0.002
Sacramento River Deep Water Channel, Light 55	307	17.2	7.73	10.3	15.8	0.08	0.001
Sacramento River across from Sherman Lake (704)	206	15.1	7.59	10.6	8.6	0.12	0.001
San Joaquin River, West of Oulton Point (812)	205	15.3	7.40	10.5	8.4	0.03	0.000
Field Duplicate: Sacramento River across from Sherman Lake (704)	206	15.1	7.59	10.6	10.0	0.13	0.001

Table A65-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 3/17/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 3/16/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	340	22.4	24.0	7.2	8.5	7.82	7.98	108	60	-
DIEPAMHR + organic matter	355	22.6	23.9	7.0	8.6	7.62	8.11	108	60	-
Old River, western arm at railroad bridge (902)	244	22.8	24.1	7.0	8.7	7.62	7.82	72	76	0.000
Old River at mouth of Holland Cut (915)	245	22.7	23.9	6.9	8.6	7.60	7.79	76	72	0.000
San Joaquin River between Hog and Turner Cut (910)	450	22.8	24.2	6.9	8.7	7.61	7.87	116	86	0.001
Sacramento River at tip of Grand Island (711)	173	22.9	24.2	7.0	8.8	7.63	7.82	60	72	0.010
Sacramento River Deep Water Channel, Light 55	301	22.9	24.2	7.1	8.7	7.82	8.05	76	80	0.004
Sacramento River across from Sherman Lake (704)	199	22.8	24.2	7.0	8.8	7.69	7.95	76	114	0.005
San Joaquin River, West of Oulton Point (812)	210	22.8	23.9	7.1	8.5	7.59	8.03	72	72	0.002
Field Duplicate: Sacramento River across from Sherman Lake (704)	204	22.7	24.5	6.9	8.7	7.71	7.94	72	82	0.006
DIEPAMHR + organic matter + 25 ppb PBO	352	23.0	24.5	7.0	8.5	7.52	8.02	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	241	23.1	24.6	6.9	8.9	7.60	7.99	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	250	22.9	24.1	6.8	8.7	7.58	7.84	-	-	-
San Joaquin River between Hog and Turner Cut (910) + 25 ppb PBO	466	22.9	24.6	6.9	8.5	7.61	7.86	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	173	23.0	24.2	7.0	8.5	7.61	8.08	-	-	-
Sacramento River Deep Water Channel, Light 55 + 25 ppb PBO	317	23.1	24.0	6.8	8.5	7.72	7.99	-	-	-
Sacramento River across from Sherman Lake (704) + 25 ppb PBO	200	22.9	24.5	7.2	8.3	7.74	8.03	-	-	-
San Joaquin River, West of Oulton Point (812) + 25 ppb PBO	212	22.9	24.3	7.0	8.5	7.66	7.94	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A66-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 3/29/07 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 3/28/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	93	2.4	-	-	NS
DIEPAMHR + organic matter	98	2.5	95	2.9	NS
High EC Control @ 16.86 mS + organic matter	86	7.0	87	8.2	NS
Carquinez Strait, West of Benicia army dock (405) ³	95	2.9	89	4.1	NS
Suisun Bay off Chipps Island (508)	100	0.0	100	0.0	NS
Grizzly Bay at Dolphin (602)	100	0.0	98	2.5	NS
Middle of Broad Slough, West end (804)	100	0.0	98	2.5	NS
Suisun Bay, East of middle point (504)	100	0.0	98	2.5	NS
Montezuma Slough at Nurse Slough (609)	100	0.0	100	0.0	NS

	MSD	PMSD
One-way ANOVA	11.6	11.9
Two-way ANOVA	16.1	16.5

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.048	0.005	-	-	NS
DIEPAMHR + organic matter	0.078	0.003	0.093	0.006	NS
High EC Control @ 16.86 mS + organic matter	0.034	0.002	0.035	0.004	NS
Carquinez Strait, West of Benicia army dock (405) ³	0.055	0.004	0.072	0.009	NS
Suisun Bay off Chipps Island (508)	0.081	0.004	0.089	0.013	NS
Grizzly Bay at Dolphin (602)	0.080	0.003	0.079	0.011	NS
Middle of Broad Slough, West end (804)	0.087	0.009	0.081	0.007	NS
Suisun Bay, East of middle point (504)	0.084	0.007	0.079	0.005	NS
Montezuma Slough at Nurse Slough (609)	0.085	0.008	0.069	0.007	NS

	MSD	PMSD
One-way ANOVA	0.026	33.3
Two-way ANOVA	0.036	46.1

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.
Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable
3. This high conductivity sample was compared to the High EC control.

Table A66-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 3/28/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Carquinez Strait, West of Benicia army dock (405)	14960	14.3	7.74	10.3	12.2	0.12	0.001
Suisun Bay off Chipps Island (508)	833	14.8	7.71	10.3	15.5	0.08	0.001
Grizzly Bay at Dolphin (602)	7240	14.3	7.81	10.6	31.7	0.08	0.001
Middle of Broad Slough, West end (804)	280	14.3	7.54	10.1	9.5	0.05	0.000
Suisun Bay, East of middle point (504)	2454	14.3	7.81	10.4	21.2	0.08	0.001
Montezuma Slough at Nurse Slough (609)	3314	15.4	7.47	8.8	83.5	0.16	0.001

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Table A66-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 3/29/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 3/28/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	347	23.3	23.5	7.1	8.6	7.69	8.04	116	60	-
DIEPAMHR + organic matter	362	23.2	23.7	6.9	8.4	7.59	8.00	116	60	-
High EC Control @ 16.86 mS + organic matter	15950	23.3	23.8	7.1	8.4	7.59	7.84	-	-	-
Carquinez Strait, West of Benicia army dock (405)	14650	23.2	23.9	7.2	8.4	7.63	7.70	1680	100	0.002
Suisun Bay off Chipps Island (508)	1061	23.3	23.9	7.0	8.7	7.74	8.08	148	72	0.004
Grizzly Bay at Dolphin (602)	3288	23.2	23.8	7.2	8.5	7.66	7.76	736	78	0.002
Middle of Broad Slough, West end (804)	353	23.1	23.8	7.1	8.7	7.33	8.14	80	72	0.003
Suisun Bay, East of middle point (504)	2534	23.2	23.6	7.2	8.5	7.70	7.81	304	72	0.002
Montezuma Slough at Nurse Slough (609)	3047	23.3	23.7	7.1	8.6	7.68	7.75	384	76	0.004
DIEPAMHR + organic matter + 25 ppb PBO	364	23.0	23.2	7.1	8.4	7.74	8.03	-	-	-
High EC Control @ 16.86mS + organic matter + 25 ppb PBO	15880	22.7	23.3	6.9	8.4	7.54	7.80	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	14180	23.3	23.3	7.3	8.4	7.67	7.81	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	1052	23.0	23.3	7.0	8.7	7.75	8.14	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	6290	23.1	23.4	7.2	8.5	7.63	7.80	-	-	-
Middle of Broad Slough, West end (804) + 25 ppb PBO	356	23.3	23.4	7.1	8.4	7.71	8.19	-	-	-
Suisun Bay, East of middle point (504) + 25 ppb PBO	2551	23.3	23.9	7.1	8.6	7.64	7.89	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	3092	23.5	23.8	7.1	8.7	7.66	7.78	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A67-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 03/30/2007 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 3/28/07 - 3/29/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	93	4.4	-	-	NA
DIEPAMHR + organic matter	98	2.3	100	0.0	NS
Napa River at Riverside Blvd. Terminus (340)	100	0.0	100	0.0	NS
Old River, western arm at railroad bridge (902)	98	2.1	100	0.0	NS
Old River at Holland Cut (915)	98	2.3	100	0.0	NS
San Joaquin River between Hog and Turner Cuts (910)	94	3.2	95	4.5	NS
San Joaquin River, West of Oulton Point (812)	98	2.5	95	2.9	NS
Sacramento River at tip of Grand Island (711)	95	3.1	90	3.7	NS
Sacramento River Deep Water Channel, Light 55	100	0.0	100	0.0	NS
Sacramento River across from Sherman Lake (704)	93	2.4	84	8.6	NS
Field Duplicate: San Joaquin R. between Hog and Turner Cuts (910)	98	2.5	-	-	NA

	MSD	PMSD
One-way ANOVA	12.6	12.9
Two-way ANOVA	15.6	16.0

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.055	0.004	-	-	NA
DIEPAMHR + organic matter	0.089	0.007	0.086	0.010	NS
Napa River at Riverside Blvd. Terminus (340)	0.097	0.005	0.079	0.002	NS
Old River, western arm at railroad bridge (902)	0.082	0.010	0.095	0.009	NS
Old River at Holland Cut (915)	0.084	0.014	0.081	0.014	NS
San Joaquin River between Hog and Turner Cuts (910)	0.087	0.009	0.069	0.007	NS
San Joaquin River, West of Oulton Point (812)	0.083	0.005	0.078	0.011	NS
Sacramento River at tip of Grand Island (711)	0.085	0.005	0.061	0.004	NS
Sacramento River Deep Water Channel, Light 55	0.092	0.008	0.078	0.008	NS
Sacramento River across from Sherman Lake (704)	0.053	0.007	0.083	0.009	NS
Field Duplicate: San Joaquin R. between Hog and Turner Cuts (910)	0.085	0.007	-	-	NA

	MSD	PMSD
One-way ANOVA	0.039	43.6
Two-way ANOVA	0.045	50.8

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable

Table A67-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 3/28/07 - 3/29/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Napa River at Riverside Blvd. Terminus (340)	3094	18.4	7.77	9.8	26.6	0.04	0.001
Old River, western arm at railroad bridge (902)	226	13.4	7.47	10.3	8.5	0.02	0.000
Old River at Holland Cut (915)	229	14.5	7.54	10.1	5.8	0.03	0.000
San Joaquin River between Hog and Turner Cuts (910)	635	15.2	7.62	9.3	4.6	0.15	0.002
San Joaquin River, West of Oulton Point (812)	201	14.6	7.54	10.0	6.7	0.06	0.001
Sacramento River at tip of Grand Island (711)	157	14.5	7.61	10.1	3.5	0.11	0.001
Sacramento River Deep Water Channel, Light 55	358	15.0	7.93	9.9	17.9	0.08	0.002
Sacramento River across from Sherman Lake (704)	201	14.7	7.72	9.8	9.3	0.00	0.000
Field Duplicate: San Joaquin R. between Hog and Turner Cuts (910)	-	-	-	-	6.4	0.15	-

Table A67-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 3/30/2007 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 3/28/07 - 3/29/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	365	23.2	23.9	6.7	8.5	7.58	8.22	116	60	-
DIEPAMHR + organic matter	376	23.4	24.4	6.7	8.4	7.47	8.07	116	60	-
Napa River at Riverside Blvd. Terminus (340)	3078	23.4	24.5	7.0	8.6	7.68	7.85	388	110	0.001
Old River, western arm at railroad bridge (902)	243	23.4	24.5	6.7	8.8	7.57	7.94	72	68	0.001
Old River at Holland Cut (915)	237	23.5	24.4	6.6	8.8	7.53	8.05	72	68	0.002
San Joaquin River between Hog and Turner Cuts (910)	625	23.6	24.0	6.9	8.7	7.69	7.95	148	94	0.006
San Joaquin River, West of Oulton Point (812)	227	23.6	23.9	6.6	8.6	7.53	7.88	84	64	0.002
Sacramento River at tip of Grand Island (711)	177	23.6	23.9	6.8	8.7	7.53	7.96	60	68	0.005
Sacramento River Deep Water Channel, Light 55	367	23.2	23.6	6.8	8.7	7.69	8.07	112	106	0.004
Sacramento River across from Sherman Lake (704)	213	23.6	23.6	6.6	8.9	7.60	7.98	72	74	0.000
Field Duplicate: San Joaquin R. between Hog and Turner Cuts (910)	612	23.5	23.6	7.0	8.6	7.64	7.98	144	92	0.006
DIEAPMHR + organic matter + 25 ppb PBO	356	23.4	23.5	6.6	8.4	7.45	7.97	-	-	-
Napa River at Riverside Blvd. Terminus (340) + 25 ppb PBO	3014	23.4	23.6	7.0	8.8	7.68	7.90	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	249	23.4	23.6	6.9	8.5	7.49	8.05	-	-	-
Old River at Holland Cut (915) + 25 ppb PBO	237	23.3	23.6	6.9	8.6	7.55	8.07	-	-	-
San Joaquin River between Hog and Turner Cuts (910) + 25 ppb PBO	625	23.6	23.6	6.8	8.6	7.66	7.98	-	-	-
San Joaquin River, West of Oulton Point (812) + 25 ppb PBO	211	23.3	23.6	6.9	8.7	7.54	8.00	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	166	23.2	23.6	6.9	8.5	7.55	8.09	-	-	-
Sacramento River Deep Water Channel, Light 55 + 25 ppb PBO	361	23.3	23.6	6.9	8.5	7.65	8.10	-	-	-
Sacramento River across from Sherman Lake (704) + 25 ppb PBO	210	23.3	23.6	6.9	8.7	7.62	8.02	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A68-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 4/13/2007 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 4/11/07 - 4/12/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	92	2.7	-	-	NA
DIEMPAMHR + organic matter	100	0.0	84	7.8	NS
Napa River at Riverside Blvd terminus (340)	100	0.0	98	2.5	NS
Old River, western arm at railroad bridge (902)	94	3.3	70	18.2	NS
Old River at mouth of Holland Cut (915)	88	2.0	93	7.5	NS
San Joaquin R. between Hog and Turner Cuts (910)	97	2.8	100	0.0	NS
San Joaquin R., West of Oulton Point (812)	86	7.0	95	5.0	NS
Sacramento River Deep Water Channel, Light 55	78	4.8	59	13.3	NS
Sacramento River at tip of Grand Island (711)	63	15.9	87	3.1	NS

	MSD	PMSD
One-way ANOVA	29.9	29.9
Two-way ANOVA	40.6	40.6

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.046	0.009	-	-	NA
DIEMPAMHR + organic matter	0.066	0.009	0.054	0.003	NS
Napa River at Riverside Blvd terminus (340)	0.071	0.010	0.041	0.008	NS
Old River, western arm at railroad bridge (902)	0.100	0.010	0.060	0.015	NS
Old River at mouth of Holland Cut (915)	0.058	0.015	0.063	0.006	NS
San Joaquin R. between Hog and Turner Cuts (910)	0.077	0.006	0.074	0.009	NS
San Joaquin R., West of Oulton Point (812)	0.052	0.012	0.060	0.010	NS
Sacramento River Deep Water Channel, Light 55	0.083	0.007	0.053	0.003	NS
Sacramento River at tip of Grand Island (711)	0.084	0.014	0.052	0.008	NS

	MSD	PMSD
One-way ANOVA	0.050	76.1
Two-way ANOVA	0.050	76.1

- Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.
Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
- NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable

Table A68-2. Summary of water chemistry measurements on samples collected by the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 4/11/07 - 4/12/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Napa River at Riverside Blvd terminus (340)	5080	18.6	8.55	12.6	17.8	0.02	0.002
Old River, western arm at railroad bridge (902)	436	8.1	7.72	12.4	8.5	0.03	0.000
Old River at mouth of Holland Cut (915)	450	8.1	7.68	12.2	5.6	0.01	0.000
San Joaquin R. between Hog and Turner Cuts (910)	649	8.3	7.8	12.9	5.2	0.08	0.001
San Joaquin R., West of Oulton Point (812)	421	8.2	7.75	12.5	5.5	0.04	0.000
Sacramento River Deep Water Channel, Light 55	295	8.8	7.91	12.3	15.0	0.14	0.002
Sacramento River at tip of Grand Island (711)	195	9.2	7.76	12.4	3.4	0.31	0.003

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Table A68-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 4/13/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 4/11/07- 1/12/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	363	20.7	24.8	7.1	8.3	7.68	8.12	106	60	-
DIEMPAMHR + organic matter	366	20.7	24.0	7.0	8.3	7.82	8.19	106	60	-
Napa River at Riverside Blvd terminus (340)	5690	21.0	23.9	7.1	8.6	7.80	8.21	660	130	0.001
Old River, western arm at railroad bridge (902)	238	20.8	23.2	7.1	8.5	7.78	8.20	72	66	0.002
Old River at mouth of Holland Cut (915)	247	21.1	24.1	7.1	8.4	7.72	8.13	76	66	0.001
San Joaquin R. between Hog and Turner Cuts (910)	552	21.0	23.4	7.0	8.6	7.72	8.06	132	100	0.004
San Joaquin R., West of Oulton Point (812)	240	21.2	24.3	7.1	8.3	7.65	8.18	68	64	0.003
Sacramento River Deep Water Channel, Light 55	265	21.2	24.2	7.1	8.4	7.80	8.15	84	78	0.009
Sacramento River at tip of Grand Island (711)	159	21.4	23.2	7.1	8.3	7.80	8.07	48	60	0.016
DIEMPAMHR + organic matter + 25 ppb PBO	361	21.3	23.8	7.2	8.3	7.60	8.20	-	-	-
Napa River at Riverside Blvd terminus (340) + 25 ppb PBO	5540	21.2	24.0	6.8	8.9	7.73	8.19	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	243	21.5	24.0	7.0	8.3	7.67	8.15	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	246	21.6	24.2	7.0	8.4	7.62	8.24	-	-	-
San Joaquin R. between Hog and Turner Cuts (910) + 25 ppb PBO	562	21.6	23.9	7.0	8.6	7.89	8.01	-	-	-
San Joaquin R., West of Oulton Point (812) + 25 ppb PBO	244	21.2	23.8	7.1	8.3	7.59	8.21	-	-	-
Sacramento River Deep Water Channel, Light 55 + 25 ppb PBO	263	21.7	24.2	7.1	8.5	7.76	8.13	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	159	21.8	24.4	7.3	8.4	7.70	8.16	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A69-1. Survival of *H. azteca* in a Toxicity Identification Evaluation initiated on 5/01/07 examining an ambient water column sample collected from the Sacramento River at Grand Island (POD site 711) by the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 4/12/07.¹

Treatment	Survival (%) ²									
	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Day 8	Day 9	Day 10
DIEPAMHR	100	100	100	100	100	100	100	100	100	100
DIEPAMHR (HA) @ 48 mg/L	100	100	100	100	100	100	100	100	97	97
DIEPAMHR (HA) + MeOH @ 0.5%	97	93	93	93	93	93	93	93	93	93
DIEPAMHR (HA) + eluate addback @ 3x	100	100	97	93	93	93	93	93	93	93
DIEPAMHR (HA) + 48 mg/L EDTA	100	97	97	97	97	97	97	97	97	97
DIEPAMHR (HA) + 24 mg/L EDTA	100	100	100	100	100	100	100	100	100	100
DIEPAMHR (HA) + 12 mg/L EDTA	100	100	97	97	97	97	97	93	93	93
DIEPAMHR (HA) + 12 mg/L STS	97	77	57	50	47	47	40	40	40	40
DIEPAMHR (HA) + 6 mg/L STS	100	73	53	53	45	45	45	45	45	45
DIEPAMHR (HA) + 3 mg/L STS	100	76	73	66	62	62	62	62	62	62
DIEPAMHR (HA) + 500X Esterase	90	50	27	13	10	7	7	7	3	3
DIEPAMHR (HA) + 500X BSA	100	90	87	87	87	87	87	84	79	79
DIEPAMHR (HA) air stripped	100	100	100	100	100	100	100	100	100	100
DIEPAMHR C8 Blank	100	97	97	97	97	97	97	97	97	97
DIEPAMHR @ 15 C	100	100	100	100	100	100	100	97	97	97
POD 711	93	90	90	90	90	90	90	90	90	90
POD 711 + 48 mg/L EDTA	100	97	97	97	97	97	97	97	97	97
POD 711 + 24 mg/L EDTA	100	97	97	93	93	93	93	93	93	93
POD 711 + 12 mg/L EDTA	100	94	91	91	91	91	91	91	91	91
POD 711 + 12 mg/L STS	93	66	62	62	62	62	62	62	62	62
POD 711 + 6 mg/L STS	97	87	83	77	77	77	77	77	77	77
POD 711 + 3 mg/L STS	100	84	77	77	77	77	77	77	77	77
POD 711 + 500X Esterase	41	24	7	7	7	7	7	3	3	0
POD 711 + 500X BSA	97	93	90	82	82	82	79	79	79	79
POD 711 air stripped	100	93	88	88	88	88	88	88	88	88
POD 711 C8 Rinsate	93	87	87	87	87	87	87	87	87	87
POD 711 @ 15 C	100	100	100	100	100	100	100	93	93	93

1. C8 column solid phase extraction performed on 4/25/07.

2. Highlighted cells indicate less than 50% survival of test organisms.

Table A70-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 4/18/2007 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 4/11/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	95	5.0	-	-	NA
DIEPAMHR + organic matter	100	0.0	98	2.5	NS
High EC Control @ 20.0 mS/cm + organic matter	78	11.1	73	9.3	NS
Middle of Broad Slough, West end (804)	100	0.0	100	0.0	NS
Suisun Bay off Chipps Island (508)	100	0.0	100	0.0	NS
Suisun Bay, East of middle point (504)	98	2.5	98	2.5	NS
Carquinez Strait, West of Benicia army dock (405) ³	87	6.3	95	2.9	NS
Grizzly Bay at Dolphin (602)	98	2.5	98	2.5	NS
Montezuma Slough at Nurse Slough (609)	100	0.0	100	0.0	NS
Sacramento River, across from Sherman Lake (704)	100	0.0	95	5.0	NS
Field Duplicate: Grizzly Bay at Dolphin (602)	100	0.0	-	-	NA

	MSD	PMSD
One-way ANOVA	20.9	20.9
Two-way ANOVA	21.7	21.7

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.058	0.006	-	-	NA
DIEPAMHR + organic matter	0.106	0.005	0.081	0.004	NS
High EC Control @ 20.0 mS/cm + organic matter	0.050	0.004	0.034	0.001	NS
Middle of Broad Slough, West end (804)	0.087	0.013	0.109	0.006	NS
Suisun Bay off Chipps Island (508)	0.103	0.003	0.099	0.005	NS
Suisun Bay, East of middle point (504)	0.100	0.005	0.077	0.005	NS
Carquinez Strait, West of Benicia army dock (405) ³	0.070	0.009	0.084	0.005	NS
Grizzly Bay at Dolphin (602)	0.110	0.007	0.117	0.011	NS
Montezuma Slough at Nurse Slough (609)	0.125	0.002	0.084	0.015	S (67%)
Sacramento River, across from Sherman Lake (704)	0.119	0.006	0.103	0.017	NS
Field Duplicate: Grizzly Bay at Dolphin (602)	0.089	0.009	-	-	NA

	MSD	PMSD
One-way ANOVA	0.034	32.2
Two-way ANOVA	0.040	37.4

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$). Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable
3. This high conductivity sample was compared to the High EC control.

Table A70-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 4/11/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Middle of Broad Slough, West end (804)	493	16.2	7.45	9.8	11.6	0.11	0.001
Suisun Bay off Chipps Island (508)	2646	15.7	7.69	10.0	22.0	0.09	0.001
Suisun Bay, East of middle point (504)	7540	15.5	7.70	9.7	14.5	0.11	0.001
Carquinez Strait, West of Benicia army dock (405)	19770	15.1	7.70	9.3	16.2	0.08	0.001
Grizzly Bay at Dolphin (602)	9050	15.6	7.80	9.6	31.6	0.06	0.001
Montezuma Slough at Nurse Slough (609)	3668	16.6	7.40	8.2	64.9	0.12	0.001
Sacramento River, across from Sherman Lake (704)	603	16.3	7.70	9.6	15.2	0.13	0.002
Field Duplicate: Grizzly Bay at Dolphin (602)	-	-	-	-	30.4	0.07	-

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Table A70-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 4/18/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 4/11/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	380	23.1	24.2	6.7	8.5	7.51	8.26	106	60	-
DIEPAMHR + organic matter	374	23.9	24.2	6.4	8.3	7.40	8.27	106	60	-
High EC Control @ 20.0 mS/cm + organic matter	19465	23.9	24.3	6.7	8.5	7.42	8.00	2200	60	-
Middle of Broad Slough, West end (804)	520	23.7	24.2	6.6	8.5	7.59	8.25	92	72	0.008
Suisun Bay off Chipps Island (508)	2484	23.9	24.2	6.5	8.5	7.50	8.06	316	74	0.004
Suisun Bay, East of middle point (504)	7285	23.9	24.2	6.6	8.9	7.49	7.85	880	90	0.003
Carquinez Strait, West of Benicia army dock (405)	19345	24.3	24.3	6.0	8.5	7.40	7.60	2240	120	0.001
Grizzly Bay at Dolphin (602)	8915	24.1	24.2	6.4	8.6	7.49	7.89	510	100	0.002
Montezuma Slough at Nurse Slough (609)	3609	23.6	24.2	6.8	8.7	7.54	8.01	440	90	0.005
Sacramento River, across from Sherman Lake (704)	635	24.2	24.2	6.5	8.3	7.53	8.21	112	70	0.009
Field Duplicate: Grizzly Bay at Dolphin (602)	9190	24.0	24.2	6.9	8.8	7.55	7.83	1060	100	0.002
DIEPAMHR + organics + 25 ppb PBO	376	23.1	24.1	6.7	8.3	7.50	8.22	-	-	-
High EC Control @ 20.0 mS/cm + organic matter + 25 ppb PBO	19295	23.8	24.2	7.0	8.7	7.48	7.84	-	-	-
Middle of Broad Slough, West end (804) + 25 ppb PBO	537	23.6	24.1	6.7	8.5	7.62	8.31	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	2610	23.4	24.2	6.5	8.9	7.04	7.74	-	-	-
Suisun Bay, East of middle point (504) + 25 ppb PBO	7275	23.4	24.1	6.8	8.3	7.54	7.81	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	18980	23.2	24.2	7.0	8.8	7.55	7.72	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	8910	23.0	24.3	6.9	8.3	7.59	8.00	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	3578	23.0	24.4	6.8	8.3	7.56	7.89	-	-	-
Sacramento River, across from Sherman Lake (704) + 25 ppb PBO	621	22.9	24.3	6.6	8.3	7.55	8.28	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A71-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 4/26/2007 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 4/25/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO ²
DIEPAMHR 042507	100	0.0	-	-	NA
DIEPAMHR + organic matter	100	0.0	96	3.7	NS
Old River, western arm at railroad bridge (902)	98	2.5	93	2.4	NS
Old River at mouth of Holland Cut (915)	100	0.0	95	5.0	NS
San Joaquin R., between Hog and Turner Cut (910)	93	4.4	100	0.0	NS
San Joaquin R., West of Oulton Point (812)	97	2.8	100	0.0	NS
Sacramento River Deep Water Channel, Light 55	95	2.9	93	3.3	NS
Sacramento River at tip of Grand Island (711)	94	5.6	92	4.8	NS
Sacramento River across from Sherman Lake (704)	95	2.9	95	5.0	NS
Middle of Broad Slough, West end (804)	95	2.9	93	4.8	NS
Field Duplicate: Middle of Broad Slough, West end (804)	95	2.8	-	-	NA

	MSD	PMSD
One-way ANOVA	14.8	14.8
Two-way ANOVA	18.2	18.5

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO ²
DIEPAMHR 042507	0.039	0.005	-	-	NA
DIEPAMHR + organic matter	0.068	0.009	0.075	0.008	NS
Old River, western arm at railroad bridge (902)	0.081	0.012	0.105	0.005	NS
Old River at mouth of Holland Cut (915)	0.089	0.012	0.102	0.006	NS
San Joaquin R., between Hog and Turner Cut (910)	0.082	0.006	0.101	0.010	NS
San Joaquin R., West of Oulton Point (812)	0.076	0.012	0.074	0.016	NS
Sacramento River Deep Water Channel, Light 55	0.071	0.009	0.078	0.013	NS
Sacramento River at tip of Grand Island (711)	0.075	0.010	0.039	0.008	NS
Sacramento River across from Sherman Lake (704)	0.099	0.009	0.054	0.004	NS
Middle of Broad Slough, West end (804)	0.082	0.007	0.077	0.005	NS
Field Duplicate: Middle of Broad Slough, West end (804)	0.076	0.012	-	-	NA

	MSD	PMSD
One-way ANOVA	0.048	70.3
Two-way ANOVA	0.049	68.6

- Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$). Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
- NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable

Table A71-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 4/25/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Old River, western arm at railroad bridge (902)	415	16.7	7.99	10.1	6.2	0.06	0.002
Old River at mouth of Holland Cut (915)	482	17.2	7.80	9.9	6.1	0.08	0.001
San Joaquin R., between Hog and Turner Cut (910)	680	17.1	8.15	11.1	10.9	0.1	0.004
San Joaquin R., West of Oulton Point (812)	298	16.2	7.88	10.6	7.3	0.1	0.002
Sacramento River Deep Water Channel, Light 55	230	16.7	7.89	9.8	19.2	0.21	0.005
Sacramento River at tip of Grand Island (711)	150	16.4	7.70	10.3	4.4	0.36	0.005
Sacramento River across from Sherman Lake (704)	357	16.1	7.90	10.0	14.5	0.16	0.003
Middle of Broad Slough, West end (804)	522	16.7	7.90	10.1	13.6	0.14	0.003
Field Duplicate: Middle of Broad Slough, West end (804)	522	16.7	7.90	10.1	12.1	0.15	0.003

Table A71-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 4/26/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 4/25/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	375	23.2	24.5	7.2	8.8	7.74	8.18	104	58	-
DIEPAMHR + organic matter	367	23.1	24.8	6.7	8.2	7.60	8.20	104	58	-
Old River, western arm at railroad bridge (902)	247	23.1	24.8	6.8	8.7	7.63	8.05	68	64	0.003
Old River at mouth of Holland Cut (915)	288	23.1	24.6	6.5	8.5	7.75	7.98	76	68	0.004
San Joaquin R., between Hog and Turner Cut (910)	621	23.0	24.8	6.9	8.7	7.83	8.00	144	88	0.005
San Joaquin R., West of Oulton Point (812)	233	23.1	24.8	6.8	8.2	7.66	8.04	60	64	0.005
Sacramento River Deep Water Channel, Light 55	242	23.0	24.9	6.2	8.8	7.76	8.01	84	82	0.011
Sacramento River at tip of Grand Island (711)	168	23.0	24.9	6.6	8.2	7.64	7.96	52	58	0.017
Sacramento River across from Sherman Lake (704)	341	23.1	24.8	6.9	8.6	7.63	8.17	76	68	0.011
Middle of Broad Slough, West end (804)	532	23.0	24.9	6.7	8.7	7.69	8.00	104	70	0.007
Field Duplicate: Middle of Broad Slough, West end (804)	543	23.0	24.8	6.7	8.9	7.64	7.89	92	70	0.006
DIEPAMHR + organic matter + 25 ppb PBO	360	23.0	23.8	6.7	8.2	7.66	8.20	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	247	23.0	24.1	6.7	8.5	7.63	8.16	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	271	23.0	24.2	6.5	8.6	7.67	8.03	-	-	-
San Joaquin R., between Hog and Turner Cut (910) + 25 ppb PBO	604	22.9	24.0	6.4	8.7	7.83	8.13	-	-	-
San Joaquin R., West of Oulton Point (812) + 25 ppb PBO	223	23.0	23.9	6.6	8.5	7.71	8.13	-	-	-
Sacramento River Deep Water Channel, Light 55 + 25 ppb PBO	244	22.9	23.9	6.7	8.3	7.79	8.15	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	161	23.0	23.7	6.6	8.8	7.70	8.17	-	-	-
Sacramento River across from Sherman Lake (704) + 25 ppb PBO	332	23.0	23.6	6.7	8.7	7.68	8.21	-	-	-
Middle of Broad Slough, West end (804) + 25 ppb PBO	516	22.9	23.7	6.8	8.8	7.68	8.00	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A72-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 4/28/2007 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 4/26/07 - 4/27/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	94	3.3	-	-	NA
DIEPAMHR + organic matter	92	2.6	95	2.9	NS
High EC Control @ 13.0 mS/cm + organic matter	98	2.5	64	11.7	S (65%)
Napa River at Riverside Blvd terminus (340)	98	2.5	93	4.8	NS
Suisun Bay off Chipps Island (508)	97	3.1	100	0.0	NS
Suisun Bay, East of middle point (504)	100	0.0	100	0.0	NS
Carquinez Strait, West of Benicia army dock (405) ³	98	2.3	95	3.1	NS
Grizzly Bay at Dolphin (602)	100	0.0	100	0.0	NS
Montezuma Slough at Nurse Slough (609)	100	0.0	98	2.5	NS

	MSD	PMSD
One-way ANOVA	10.7	11.6
Two-way ANOVA	18.5	19.7

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.039	0.007	-	-	NA
DIEPAMHR + organic matter	0.038	0.008	0.061	0.004	NS
High EC Control @ 13.0 mS/cm + organic matter	0.024	0.006	0.019	0.003	NS
Napa River at Riverside Blvd terminus (340)	0.046	0.004	0.049	0.012	NS
Suisun Bay off Chipps Island (508)	0.040	0.010	0.068	0.004	NS
Suisun Bay, East of middle point (504)	0.049	0.004	0.051	0.005	NS
Carquinez Strait, West of Benicia army dock (405) ³	0.062	0.007	0.035	0.004	NS
Grizzly Bay at Dolphin (602)	0.069	0.002	0.093	0.009	NS
Montezuma Slough at Nurse Slough (609)	0.090	0.006	0.099	0.005	NS

	MSD	PMSD
One-way ANOVA	0.031	82.6
Two-way ANOVA	0.032	65.0

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$). Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable
3. This high conductivity sample was compared to the High EC control.

Table A72-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 4/26/07 - 4/27/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Napa River at Riverside Blvd terminus (340)	7130	16.5	7.70	9.1	23.3	0.12	0.001
Suisun Bay off Chipps Island (508)	786	17.1	8.01	10.1	15.4	0.11	0.003
Suisun Bay, East of middle point (504)	4428	18.4	8.05	10.1	20.3	0.13	0.004
Carquinez Strait, West of Benicia army dock (405)	12780	18.2	8.00	10.2	18.2	0.13	0.003
Grizzly Bay at Dolphin (602)	7350	19.0	8.01	10.0	28.8	0.11	0.003
Montezuma Slough at Nurse Slough (609)	4477	17.7	7.66	8.8	54.4	0.15	0.002

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Table A72-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 4/28/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 4/26/07- 4/27/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	322	23.0	24.8	6.6	8.3	7.72	8.05	104	58	-
DIEPAMHR + organic matter	368	22.9	24.9	6.9	8.2	7.65	8.08	104	58	-
High EC Control @ 13.0 mS/cm + organic matter	13015	22.9	24.9	6.9	8.7	7.67	7.84	444	92	-
Napa River at Riverside Blvd terminus (340)	7565	22.9	24.8	6.6	8.7	7.83	7.89	184	116	0.004
Suisun Bay off Chipps Island (508)	707	23.1	24.7	6.2	8.6	7.72	7.86	-	-	0.005
Suisun Bay, East of middle point (504)	4344	23.0	24.9	7.0	8.5	7.64	7.83	-	-	0.005
Carquinez Strait, West of Benicia army dock (405)	12820	22.9	24.9	6.2	8.8	7.64	7.71	-	-	0.003
Grizzly Bay at Dolphin (602)	7435	22.9	24.7	6.5	8.8	7.68	7.83	-	-	0.004
Montezuma Slough at Nurse Slough (609)	4438	23.0	24.9	6.9	8.9	7.70	7.85	-	-	0.005
DIEPAMHR + organic matter + 25 ppb PBO	367	23.1	24.8	6.8	8.3	7.64	8.12	-	-	-
High EC Control @ 13.0 mS/cm + organic matter + 25 ppb PBO	12655	22.9	24.9	7.0	8.5	7.65	7.86	-	-	-
Napa River at Riverside Blvd terminus (340) + 25 ppb PBO	7355	22.8	24.7	6.9	8.6	7.84	7.90	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	721	22.8	24.9	7.0	8.4	7.82	8.16	-	-	-
Suisun Bay, East of middle point (504) + 25 ppb PBO	4300	22.9	24.8	7.0	8.7	7.63	7.93	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	12735	22.8	24.9	6.9	8.7	7.66	7.78	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	7320	22.9	24.9	6.8	8.8	7.65	7.84	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	4331	22.8	24.8	7.0	8.6	7.68	7.88	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A73-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 05/10/2007 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 5/08/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	100	0.0	-	-	NA
DIEPAMHR + organic matter	97	3.1	97	2.8	NS
High EC Control @ 10.0 mS/cm	79	18.0	100	0.0	NS
Sacramento Deep Water Channel, Light 55	97	2.8	100	0.0	NS
Sacramento R. across from Sherman Lake (704)	100	0.0	100	0.0	NS
Old River, western arm at railroad bridge (902)	100	0.0	92	2.7	NS
San Joaquin River between Hog and Turner Cuts (910)	93	7.5	100	0.0	NS
Middle of Broad Slough, West end (804)	98	2.5	95	5.0	NS
San Joaquin River, West of Oulton Point (812)	97	2.8	95	2.9	NS
Old River at mouth of Holland Cut (915)	95	2.9	98	2.5	NS
Sacramento River at tip of Grand Island (711)	93	4.8	98	2.5	NS
Napa River at Riverside Blvd. terminus (340) ³	100	0.0	98	2.5	NS

	MSD	PMSD
One-way ANOVA	30.1	31.1
Two-way ANOVA	25.6	26.3

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.048	0.004	-	-	NA
DIEPAMHR + organic matter	0.094	0.004	0.035	0.006	S (37%)
High EC Control @ 10.0 mS/cm	0.069	0.004	0.041	0.007	NS
Sacramento Deep Water Channel, Light 55	0.075	0.006	0.091	0.007	NS
Sacramento R. across from Sherman Lake (704)	0.103	0.012	0.117	0.009	NS
Old River, western arm at railroad bridge (902)	0.076	0.017	0.085	0.005	NS
San Joaquin River between Hog and Turner Cuts (910)	0.072	0.008	0.106	0.002	NS
Middle of Broad Slough, West end (804)	0.075	0.007	0.096	0.016	NS
San Joaquin River, West of Oulton Point (812)	0.085	0.012	0.082	0.012	NS
Old River at mouth of Holland Cut (915)	0.081	0.007	0.087	0.009	NS
Sacramento River at tip of Grand Island (711)	0.061	0.006	0.061	0.005	NS
Napa River at Riverside Blvd. terminus (340) ³	0.059	0.009	0.069	0.003	NS

	MSD	PMSD
One-way ANOVA	0.044	46.7
Two-way ANOVA	0.046	49.3

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable

3. This high conductivity sample was compared to the High EC control.

Table A73-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 5/08/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total	Unionized
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)		Ammonia Nitrogen (mg/L)	Ammonia (mg/L)
Sacramento Deep Water Channel, Light 55	214	19.9	8.20	8.7	12.1	0.220	0.012
Sacramento R. across from Sherman Lake (704)	771	18.1	8.00	9.4	16.1	0.000	0.000
Old River, western arm at railroad bridge (902)	252	19.8	8.16	8.8	7.7	0.010	0.001
San Joaquin River between Hog and Turner Cuts (910)	462	20.5	8.18	8.7	6.5	0.090	0.005
Middle of Broad Slough, West end (804)	646	18.5	8.20	9.3	10.7	0.070	0.003
San Joaquin River, West of Oulton Point (812)	227	18.7	8.32	9.2	7.6	0.000	0.000
Old River at mouth of Holland Cut (915)	277	21.0	8.17	8.4	3.9	0.010	0.001
Sacramento River at tip of Grand Island (711)	178	19.4	8.05	8.6	5.6	0.230	0.009
Napa River ar Riverside Blvd. terminus (340)	9640	22.8	8.26	13.1	19.5	0.000	0.000

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Table A73-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 5/10/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 5/08/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	357	24.6	24.8	6.3	8.4	7.66	8.00	72	80	-
DIEPAMHR +organic matter	358	24.7	24.8	6.0	8.2	7.53	8.11	72	80	-
High EC Control @ 10.0 mS/cm + organic matter	9580	24.1	24.9	6.2	8.3	7.37	7.72	-	-	-
Sacramento Deep Water Channel, Light 55	232	24.7	24.7	5.9	8.9	7.80	7.96	72	76	0.010
Sacramento R. across from Sherman Lake (704)	801	24.4	24.8	6.4	8.6	7.47	7.87	124	68	0.000
Old River, western arm at railroad bridge (902)	270	24.7	24.8	5.9	8.8	7.51	7.97	80	64	0.000
San Joaquin River between Hog and Turner Cuts (910)	468	24.7	24.9	6.5	8.8	7.55	7.77	104	68	0.002
Middle of Broad Slough, West end (804)	658	24.8	24.8	6.5	8.6	7.56	7.80	108	68	0.002
San Joaquin River, West of Oulton Point (812)	239	24.8	24.8	6.3	8.5	7.59	7.92	72	64	0.000
Old River at mouth of Holland Cut (915)	299	24.8	24.9	6.2	8.9	7.54	7.76	80	66	0.000
Sacramento River at tip of Grand Island (711)	194	24.6	24.9	6.4	8.2	7.56	8.03	68	68	0.012
Napa River ar Riverside Blvd. terminus (340)	9035	24.7	24.9	6.2	8.6	7.51	7.90	356	122	0.000
DIEPAMHR +organic matter + 25 ppb PBO	357	24.2	24.9	6.5	8.2	7.67	7.81	-	-	-
High EC Control @ 10.0 mS/cm + organic matter + 25 ppb PBO	9610	24.1	25.0	6.5	8.4	7.42	7.72	-	-	-
Sacramento Deep Water Channel, Light 55 + 25 ppb PBO	236	24.2	24.7	6.4	8.4	7.74	8.07	-	-	-
Sacramento R. across from Sherman Lake (704) + 25 ppb PBO	789	24.2	24.7	6.5	8.3	7.52	7.94	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	274	24.3	24.7	6.2	8.4	7.62	7.92	-	-	-
San Joaquin River between Hog and Turner Cuts (910) + 25 ppb PBO	468	24.3	24.6	6.4	8.8	7.58	7.81	-	-	-
Middle of Broad Slough, West end (804) + 25 ppb PBO	652	24.0	24.7	6.6	8.7	7.57	7.92	-	-	-
San Joaquin River, West of Oulton Point (812) + 25 ppb PBO	242	24.4	24.7	6.3	8.8	7.49	7.97	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	294	24.1	24.8	6.3	8.6	7.48	7.95	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	196	24.3	24.8	6.3	8.6	7.49	7.98	-	-	-
Napa River ar Riverside Blvd. terminus (340) + 25 ppb PBO	9035	24.4	24.7	6.2	8.8	7.61	8.01	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A74-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 5/11/2007 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 5/10/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	97	2.8	-	-	NA
DIEPAMHR + organic matter	87	1.9	97	2.8	NS
High EC control @ 12.79 mS/cm + organic matter	85	8.6	79	8.2	NS
High EC Control @ 22.5 mS/cm + organic matter	57	14.5	66	6.1	NS
Suisun Bay off Chipps Island (508)	100	0.0	92	5.3	NS
Suisun Bay, East of middle point (504)	92	2.7	98	2.5	NS
Carquinez Strait, West of Benicia army dock (405) ³	85	5.7	57	7.7	NS
Grizzly Bay at Dolphin (602) ⁴	93	4.5	100	0.0	NS
Montezuma Slough at Nurse Slough (609)	100	0.0	98	2.5	NS
Field Duplicate: Suisun Bay, East of middle point (504)	83	6.9	-	-	NS

	MSD	PMSD
One-way ANOVA	30.6	31.5
Two-way ANOVA	29.6	30.5

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.064	0.010	-	-	NA
DIEPAMHR + organic matter	0.089	0.013	0.068	0.004	NS
High EC control @ 12.79 mS/cm + organic matter	0.079	0.011	0.052	0.008	NS
High EC Control @ 22.5 mS/cm + organic matter	0.059	0.011	0.040	0.004	NS
Suisun Bay off Chipps Island (508)	0.084	0.006	0.076	0.006	NS
Suisun Bay, East of middle point (504)	0.054	0.002	0.044	0.004	NS
Carquinez Strait, West of Benicia army dock (405) ³	0.042	0.002	0.023	0.004	NS
Grizzly Bay at Dolphin (602) ⁴	0.076	0.007	0.058	0.006	NS
Montezuma Slough at Nurse Slough (609)	0.074	0.010	0.043	0.007	NS
Field Duplicate: Suisun Bay, East of middle point (504)	0.075	0.008	-	-	NS

	MSD	PMSD
One-way ANOVA	0.042	47.4
Two-way ANOVA	0.037	41.5

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable

3. This high conductivity sample was compared to the 22.5 mS/cm High EC control.

4. This high conductivity sample was compared to the 12.79 mS/cm High EC control.

Table A74-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 5/10/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Suisun Bay off Chipps Island (508)	3710	17.4	8.04	9.6	18.5	0.08	0.002
Suisun Bay, East of middle point (504)	6500	17.6	8.01	8.5	11.0	0.09	0.002
Carquinez Strait, West of Benicia army dock (405)	21870	15.8	7.92	9.7	7.6	0.09	0.001
Grizzly Bay at Dolphin (602)	12300	16.8	8.03	9.6	54.4	0.10	0.002
Montezuma Slough at Nurse Slough (609)	4713	17.9	7.88	8.6	40.2	0.09	0.002
Field Duplicate: Suisun Bay, East of middle point (504)	6500	17.6	8.01	8.5	10.7	0.08	0.002

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Table A74-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 5/11/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 5/10/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	354	22.8	24.7	6.7	8.3	7.54	8.05	72	80	-
DIEPAMHR + organic matter	350	22.9	25.4	6.9	8.5	7.45	8.06	72	80	-
High EC control @ 12.79 mS/cm + organic matter	12140	22.8	25.0	7.2	8.6	7.45	7.90	1640	100	-
High EC Control @ 22.5 mS/cm + organic matter	21475	22.8	25.1	6.6	8.2	7.47	7.69	2640	120	-
Suisun Bay off Chipps Island (508)	4034	22.8	25.1	6.9	8.8	7.54	7.93	428	72	0.003
Suisun Bay, East of middle point (504)	7990	22.7	24.8	6.9	8.6	7.52	7.85	900	100	0.003
Carquinez Strait, West of Benicia army dock (405)	21145	22.8	24.9	6.9	8.1	7.55	7.76	2640	140	0.002
Grizzly Bay at Dolphin (602)	11900	22.9	24.7	7.0	8.3	7.54	7.80	1720	100	0.002
Montezuma Slough at Nurse Slough (609)	4718	22.9	24.6	7.0	8.4	7.62	7.84	540	88	0.003
Field Duplicate: Suisun Bay, East of middle point (504)	7980	22.8	25.0	7.1	8.4	7.54	7.84	868	90	0.002
DIEPAMHR + organic matter + 25 ppb PBO	378	22.8	24.7	6.9	8.3	7.57	8.09	-	-	-
High EC control @ 12.79 mS/cm + organic matter + 25 ppb PBO	12105	22.8	25.0	6.8	8.8	7.46	7.93	-	-	-
High EC Control @ 22.5 mS/cm + organic matter + 25 ppb PBO	21535	22.8	24.6	6.9	8.3	7.53	7.89	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	3748	22.7	24.7	6.8	8.7	7.53	7.90	-	-	-
Suisun Bay, East of middle point (504) + 25 ppb PBO	8030	22.6	24.6	7.1	8.8	7.51	7.84	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	21300	22.6	24.6	6.7	8.2	7.56	7.76	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	11835	23.0	24.6	7.0	8.7	7.55	7.86	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	4589	22.6	24.6	7.2	8.8	7.62	7.80	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A75-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 05/24/2007 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 5/22/07 - 5/23/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	90	7.1	100	0.0	NS
DIEPAMHR + organic matter	92	2.6	98	2.5	NS
High EC Control @ 13.55 mS/cm + organic matter	93	4.8	90	4.1	NS
High EC Control @ 17.00 mS/cm + organic matter	85	6.4	84	5.7	NS
Sacramento R. Deep Water Channel, Light 55	89	6.1	93	2.5	NS
Sacramento R. at tip of Grand Island (711)	66	13.3	77	4.8	NS
San Joaquin R. at Stockton WWTF	100	0.0	100	0.0	NS
Napa River at Riverside Blvd Terminus (340) ³	97	2.8	93	7.1	NS
Sacramento R. across from Sherman Lake (508)	92	2.6	98	2.3	NS
Suisun Bay, East of middle point (504)	90	10.0	98	2.5	NS
Carquinez Strait, West of Benicia army dock (405) ⁴	84	3.5	85	2.7	NS

	MSD	PMSD
One-way ANOVA	31.35	34.00
Two-way ANOVA	27.80	30.15

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.045	0.002	0.041	0.004	NS
DIEPAMHR + organic matter	0.067	0.003	0.042	0.008	NS
High EC Control @ 13.55 mS/cm + organic matter	0.036	0.005	0.036	0.009	NS
High EC Control @ 17.00 mS/cm + organic matter	0.047	0.007	0.038	0.009	NS
Sacramento R. Deep Water Channel, Light 55	0.084	0.005	0.052	0.007	NS
Sacramento R. at tip of Grand Island (711)	0.090	0.012	0.066	0.003	NS
San Joaquin R. at Stockton WWTF	0.085	0.009	0.084	0.002	NS
Napa River at Riverside Blvd Terminus (340) ³	0.054	0.002	0.046	0.012	NS
Sacramento R. across from Sherman Lake (508)	0.090	0.005	0.040	0.004	S (44%)
Suisun Bay, East of middle point (504)	0.053	0.009	0.043	0.004	NS
Carquinez Strait, West of Benicia army dock (405) ⁴	0.038	0.004	0.039	0.003	NS

	MSD	PMSD
One-way ANOVA	0.03	49.60
Two-way ANOVA	0.04	53.51

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable

3. This high conductivity sample was compared to the 13.55 mS/cm control.

4. This high conductivity sample was compared to the 17.00 mS/cm control.

Table A75-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 5/22/07 - 5/23/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Sacramento R. Deep Water Channel, Light 55	289	20.7	8.20	9.2	30.9	0.11	0.001
Sacramento R. at tip of Grand Island (711)	202	21.2	7.92	9.0	5.7	0.32	0.003
San Joaquin R. at Stockton WWTF	345	20.3	8.80	11.0	31.3	0.21	0.003
Napa River at Riverside Blvd Terminus (340)	13490	21.9	7.92	9.8	14.9	0.01	0.000
Sacramento R. across from Sherman Lake (508)	3459	18.9	8.05	9.2	15.4	0.07	0.001
Suisun Bay, East of middle point (504)	7420	18.7	8.01	9.0	13.9	0.08	0.001
Carquinez Strait, West of Benicia army dock (405)	16100	18.5	7.95	8.8	11.5	0.09	0.001

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Table A75-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 05/24/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 5/22/07- 5/23/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	356	23.3	24.0	6.8	8.2	7.57	7.90	112	62	-
DIEPAMHR + organic matter	357	23.0	23.3	6.5	8.0	7.68	7.87	112	62	-
High EC control @ 13.55 mS/cm + organic matter	13050	23.4	24.0	7.1	7.8	7.45	7.73	1520	76	-
High EC Control @ 17.00 mS/cm + organic matter	16345	23.4	24.1	7.1	7.8	7.53	7.71	1920	85	-
Sacramento R. Deep Water Channel, Light 55	231	23.1	23.4	5.5	8.4	7.64	7.79	76	78	0.002
Sacramento R. at tip of Grand Island (711)	175	23.1	23.4	5.9	8.3	7.53	7.69	56	62	0.007
San Joaquin R. at Stockton WWTP	346	23.3	24.2	6.0	8.7	7.63	7.99	80	60	0.010
Napa River at Riverside Blvd. Terminus (340)	12870	23.3	24.2	6.3	8.7	7.54	7.85	1520	160	0.000
Sacramento R. across from Sherman Lake (508)	3870	23.4	24.7	6.4	8.5	7.63	7.71	380	90	0.001
Suisun Bay, East of middle point (504)	7140	23.4	24.3	6.3	8.0	7.48	7.66	820	90	0.001
Carquinez Strait, West of Benicia army dock (405)	16515	23.4	24.2	6.5	7.8	7.48	7.67	1880	100	0.001
DIEPAMHR + 25 ppb PBO	361	23.2	23.4	6.5	8.1	7.77	7.96	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	377	23.0	23.4	6.5	8.3	7.71	7.87	-	-	-
High EC control @ 13.55 mS/cm + organic matter + 25 ppb PBO	13055	23.0	23.4	7.0	8.7	7.40	7.73	-	-	-
High EC Control @ 17.00 mS/cm + organic matter + 25 ppb PBO	16235	22.8	23.0	6.5	8.3	7.53	7.64	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	235	23.0	23.1	6.5	8.6	7.81	8.01	-	-	-
Sacramento R. at tip of Grand Island (711) + 25 ppb PBO	176	22.8	23.3	6.4	8.3	7.51	7.83	-	-	-
San Joaquin R. at Stockton WWTP + 25 ppb PBO	319	22.5	23.4	6.8	8.7	7.56	7.97	-	-	-
Napa River at Riverside Blvd. Terminus (340) + 25 ppb PBO	12565	22.4	23.4	6.7	8.6	7.63	7.70	-	-	-
Sacramento R. across from Sherman Lake (508) + 25 ppb PBO	21730	22.2	23.1	7.2	8.4	7.66	7.75	-	-	-
Suisun Bay, East of middle point (504) + 25 ppb PBO	7075	22.0	23.7	6.9	8.3	7.53	7.62	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	16515	22.0	23.7	7.0	8.4	7.49	7.61	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A76-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 05/25/2007 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 5/23/07 - 5/24/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	83	9.6	95	2.9	NS
DIEPAMHR + organic matter	92	2.8	83	13.2	NS
Middle of Broad Slough, West end (804)	98	2.5	95	2.9	NS
Grizzly Bay @ Dolphin (602)	92	4.8	95	2.9	NS
Sacramento R., across from Sherman Lake (704)	100	0.0	100	0.0	NS
Montezuma Slough @ Nurse Slough (609)	100	0.0	100	0.0	NS
Old River @ Holland Cut (902)	87	2.4	90	4.6	NS
San Joaquin R. @ Hog and Turner cuts	92	2.7	68	23.6	NS
Old River @ railroad bridge, west (915)	89	4.5	86	6.2	NS
San Joaquin R., West of Oulton Point (812)	98	2.5	89	4.1	NS
Field Duplicate: San Joaquin R., west of Oulton Point (812)	95	2.9	-	-	NA

	MSD	PMSD
One-way ANOVA	19.80	21.55
Two-way ANOVA	26.60	30.37

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.062	0.007	0.038	0.002	NS
DIEPAMHR + organic matter	0.057	0.006	0.036	0.011	NS
Middle of Broad Slough, West end (804)	0.070	0.004	0.082	0.015	NS
Grizzly Bay @ Dolphin (602)	0.054	0.005	0.046	0.007	NS
Sacramento R., across from Sherman Lake (704)	0.060	0.000	0.053	0.003	NS
Montezuma Slough @ Nurse Slough (609)	0.065	0.006	0.067	0.005	NS
Old River @ Holland Cut (902)	0.054	0.003	0.053	0.009	NS
San Joaquin R. @ Hog and Turner cuts	0.049	0.009	0.072	0.007	NS
Old River @ railroad bridge, west (915)	0.056	0.014	0.073	0.013	NS
San Joaquin R., West of Oulton Point (812)	0.067	0.007	0.068	0.003	NS
Field Duplicate: San Joaquin R., west of Oulton Point (812)	0.061	0.005	-	-	NA

	MSD	PMSD
One-way ANOVA	0.03	59.50
Two-way ANOVA	0.04	73.22

- Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.
Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
- NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable

Table A76-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 5/23/07 - 5/24/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Middle of Broad Slough, West end (804)	664	18.8	8.20	9.2	14.8	0.04	0.002
Grizzly Bay @ Dolphin (602)	8720	18.7	8.01	8.9	23.9	0.05	0.001
Sacramento R., across from Sherman Lake (704)	308	18.8	8.20	9.2	15.3	0.09	0.005
Montezuma Slough @ Nurse Slough (609)	5400	19.1	7.80	8.1	40.9	0.08	0.001
Old River @ Holland Cut (902)	303	20.0	8.19	8.7	5.5	0.02	0.001
San Joaquin R. @ Hog and Turner cuts (910)	431	20.7	7.80	8.6	6.6	0.10	0.002
Old River @ railroad bridge, west (915)	380	20.7	8.14	8.6	4.5	0.03	0.002
San Joaquin R., West of Oulton Point (812)	277	19.7	8.12	9.2	4.9	0.05	0.002
Field Duplicate: San Joaquin R., west of Oulton Point (812)	277	19.7	8.12	9.2	5.6	0.04	0.002

Table A76-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 5/25/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 5/23/07 - 5/24/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	351	22.6	24.1	6.8	8.1	7.61	8.12	112	62	-
DIEPAMHR + organic matter	355	22.7	24.7	6.6	8.1	7.53	8.00	112	62	-
Middle of Broad Slough, West end (804)	628	22.7	24.6	6.6	8.6	7.56	7.96	108	68	0.002
Grizzly Bay @ Dolphin (602)	8120	22.7	24.6	6.7	8.2	7.46	7.81	920	100	0.001
Sacramento R., across from Sherman Lake (704)	426	22.7	23.9	6.5	8.3	7.55	8.14	80	70	0.006
Montezuma Slough @ Nurse Slough (609)	4882	22.7	24.1	6.9	8.2	7.51	7.80	300	80	0.002
Old River @ Holland Cut (902)	373	22.7	24.5	6.8	8.4	7.61	7.93	76	66	0.001
San Joaquin R. @ Hog and Turner cuts (910)	382	22.7	23.8	6.5	8.3	7.53	7.96	40	80	0.004
Old River @ railroad bridge, west (915)	335	22.7	24.1	6.5	8.3	7.59	7.89	84	72	0.001
San Joaquin R., West of Oulton Point (812)	263	22.7	23.9	7.0	8.3	7.62	7.95	72	64	0.002
Field Duplicate: San Joaquin R., west of Oulton Point (812)	268	22.7	23.9	6.7	8.2	7.59	7.94	72	62	0.002
DIEPAMHR + 25 ppb PBO	354	22.7	24.0	7.1	8.2	7.63	8.15	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	349	22.7	24.2	6.4	8.1	7.61	8.07	-	-	-
Middle of Broad Slough, West end (804) + 25 ppb PBO	617	22.7	24.2	6.7	8.2	7.58	8.00	-	-	-
Grizzly Bay @ Dolphin (602) + 25 ppb PBO	7990	22.7	24.2	7.3	8.4	7.47	7.77	-	-	-
Sacramento R., across from Sherman Lake (704) + 25 ppb PBO	417	22.7	23.9	6.5	8.6	7.66	8.04	-	-	-
Montezuma Slough @ Nurse Slough (609) + 25 ppb PBO	4867	22.7	23.7	6.9	8.4	7.49	7.73	-	-	-
Old River @ Holland Cut (902) + 25 ppb PBO	385	22.7	23.5	6.6	8.2	7.64	8.06	-	-	-
San Joaquin R. @ Hog and Turner cuts (910) + 25 ppb PBO	392	22.7	23.9	6.5	8.4	7.57	7.86	-	-	-
Old River @ railroad bridge, west (915) + 25 ppb PBO	331	22.7	23.6	6.5	8.7	7.61	7.91	-	-	-
San Joaquin R., West of Oulton Point (812) + 25 ppb PBO	268	22.8	23.8	6.8	8.5	7.60	8.04	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A77-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 06/07/2007 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 6/06/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	98	2.5	100	0.0	NS
DIEPAMHR + organic matter	98	2.5	100	0.0	NS
Sacramento R. at tip of Grand Island (711)	92	2.6	98	2.5	NS
Middle of Broad Slough, West end (804)	98	2.5	100	0.0	NS
Sacramento R. across from Sherman Lake (704)	98	2.5	100	0.0	NS
Sacramento R. Deep Water Channel, Light 55	100	0.0	100	0.0	NS
Old River at mouth of Holland Cut (915)	100	0.0	98	2.5	NS
Old River, western arm at railroad bridge (902)	95	2.9	95	2.9	NS
San Joaquin R., West of Oulton Point (812)	100	0.0	95	5.0	NS
San Joaquin R., between Hog and Turner Cuts (910)	93	2.5	95	2.9	NS

	MSD	PMSD
One-way ANOVA	10.39	10.65
Two-way ANOVA	11.75	11.90

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.031	0.005	0.029	0.004	NS
DIEPAMHR + organic matter	0.051	0.006	0.048	0.007	NS
Sacramento R. at tip of Grand Island (711)	0.068	0.006	0.056	0.005	NS
Middle of Broad Slough, West end (804)	0.070	0.005	0.071	0.007	NS
Sacramento R. across from Sherman Lake (704)	0.076	0.006	0.068	0.006	NS
Sacramento R. Deep Water Channel, Light 55	0.064	0.006	0.080	0.006	NS
Old River at mouth of Holland Cut (915)	0.066	0.005	0.064	0.007	NS
Old River, western arm at railroad bridge (902)	0.045	0.005	0.115	0.006	S (255%)
San Joaquin R., West of Oulton Point (812)	0.060	0.007	0.110	0.013	S (183%)
San Joaquin R., between Hog and Turner Cuts (910)	0.061	0.010	0.104	0.011	S (170%)

	MSD	PMSD
One-way ANOVA	0.03	59.26
Two-way ANOVA	0.04	73.74

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable

Table A77-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 6/06/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Sacramento R. at tip of Grand Island (711)	205	20.1	7.88	7.7	3.4	0.410	0.011
Middle of Broad Slough, West end (804)	1627	18.1	8.24	9.2	17.8	0.090	0.004
Sacramento R. across from Sherman Lake (704)	1534	17.8	8.17	9.2	20.3	0.050	0.002
Sacramento R. Deep Water Channel, Light 55	225	18.7	7.94	8.8	26.4	0.150	0.004
Old River at mouth of Holland Cut (915)	344	20.7	8.14	7.9	4.7	0.040	0.002
Old River, western arm at railroad bridge (902)	289	19.9	8.22	8.3	13.2	0.040	0.002
San Joaquin R., West of Oulton Point (812)	259	19.1	8.33	9.1	8.3	0.050	0.003
San Joaquin R., between Hog and Turner Cuts (910)	486	21.9	7.80	8.6	8.9	0.180	0.005

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Table A77-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 06/07/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 6/06/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	347	23.0	23.4	6.5	8.4	7.64	7.93	104	60	-
DIEPAMHR + organic matter	348	23.0	23.3	6.4	8.4	7.54	7.99	104	60	-
Sacramento R. at tip of Grand Island (711)	184	23.0	23.5	6.0	8.5	7.52	7.76	60	68	0.011
Middle of Broad Slough, West end (804)	1532	23.0	23.5	6.2	8.7	7.47	7.77	196	70	0.002
Sacramento R. across from Sherman Lake (704)	1458	22.9	23.5	6.5	8.8	7.53	7.82	188	72	0.001
Sacramento R. Deep Water Channel, Light 55	205	23.0	23.4	6.0	8.4	7.59	7.93	72	74	0.006
Old River at mouth of Holland Cut (915)	338	23.0	23.4	6.2	8.6	7.57	7.73	76	70	0.001
Old River, western arm at railroad bridge (902)	281	23.2	23.4	6.3	8.6	7.56	7.94	76	66	0.002
San Joaquin R., West of Oulton Point (812)	269	23.1	23.3	6.3	8.4	7.50	8.02	68	64	0.002
San Joaquin R., between Hog and Turner Cuts (910)	449	23.3	23.3	6.1	8.2	7.58	7.73	116	76	0.004
DIEPAMHR	340	22.8	23.2	6.6	8.4	7.60	7.96	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	339	23.0	23.7	6.6	8.5	7.55	7.95	-	-	-
Sacramento R. at tip of Grand Island (711) + 25 ppb PBO	182	22.9	23.1	5.8	8.4	7.50	7.84	-	-	-
Middle of Broad Slough, West end (804) + 25 ppb PBO	1533	22.7	23.1	6.4	8.7	7.51	7.72	-	-	-
Sacramento R. across from Sherman Lake (704) + 25 ppb PBO	1458	22.8	23.3	6.4	8.6	7.57	7.78	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	223	22.8	23.2	6.0	8.5	7.60	7.89	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	338	22.9	23.5	6.3	8.9	7.59	7.86	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	267	22.7	23.3	6.1	8.6	7.58	7.82	-	-	-
San Joaquin R., West of Oulton Point (812) + 25 ppb PBO	244	22.7	23.3	6.2	8.7	7.62	7.81	-	-	-
San Joaquin R., between Hog and Turner Cuts (910) + 25 ppb PBO	441	22.9	23.5	6.1	8.3	7.57	7.64	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A78-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 06/08/2007 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 6/06/07 - 6/07/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	97	3.1	100	0.0	NS
DIEPAMHR + organic matter	95	2.9	95	2.9	NS
High EC Control @ 15.55 mS/cm	78	8.5	74	16.8	NS
High EC Control @ 21.37 mS/cm	80	8.2	72	7.3	NS
Napa River at Riverside Blvd Terminus (340) ³	98	2.5	89	4.2	NS
Suisun Bay off Chipps Island (508)	100	0.0	100	0.0	NS
Suisun Bay, East of middle point (504)	98	2.5	95	2.9	NS
Grizzly Bay at Dolphin (602)	100	0.0	100	0.0	NS
Carquinez Strait, West of Benicia army dock (405) ⁴	81	9.4	88	8.9	NS
Montezuma Slough at Nurse Slough (609)	98	2.3	95	5.0	NS
Cache Creek / Ulatris Creek Confluence	95	2.9	100	0.0	NS

	MSD	PMSD
One-way ANOVA	24.49	25.78
Two-way ANOVA	30.94	32.57

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.019	0.005	0.028	0.002	NS
DIEPAMHR + organic matter	0.040	0.004	0.037	0.005	NS
High EC Control @ 15.55 mS/cm	0.048	0.003	0.031	0.009	NS
High EC Control @ 21.37 mS/cm	0.033	0.006	0.025	0.003	NS
Napa River at Riverside Blvd Terminus (340) ³	0.031	0.008	0.032	0.004	NS
Suisun Bay off Chipps Island (508)	0.064	0.007	0.062	0.011	NS
Suisun Bay, East of middle point (504)	0.056	0.012	0.049	0.007	NS
Grizzly Bay at Dolphin (602)	0.052	0.005	0.056	0.004	NS
Carquinez Strait, West of Benicia army dock (405) ⁴	0.025	0.005	0.035	0.006	NS
Montezuma Slough at Nurse Slough (609)	0.067	0.008	0.063	0.008	NS
Cache Creek / Ulatris Creek Confluence	0.063	0.007	0.058	0.006	NS

	MSD	PMSD
One-way ANOVA	0.03	81.31
Two-way ANOVA	0.03	85.92

- Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$). Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
- NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable
- This high conductivity sample was compared to the High EC control @ 15.55 mS/cm.
- This high conductivity sample was compared to the High EC control @ 21.37 mS/cm.

Table A78-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 6/06/07 - 6/07/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Napa River at Riverside Blvd Terminus (340)	13870	20.7	7.62	8.7	6.8	0.000	0.000
Suisun Bay off Chipps Island (508)	4035	18.3	7.90	9.5	11.5	0.040	0.001
Suisun Bay, East of middle point (504)	8910	17.8	7.96	9.4	8.3	0.000	0.000
Grizzly Bay at Dolphin (602)	9140	18.2	8.09	9.7	18.4	0.000	0.000
Carquinez Strait, West of Benicia army dock (405)	21160	17.6	7.90	9.4	13.1	0.000	0.000
Montezuma Slough at Nurse Slough (609)	5570	19.4	7.77	8.5	18.9	0.040	0.001
Cache Creek / Ulatis Creek Confluence	272	20.3	7.81	8.5	27.7	0.200	0.005

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Table A78-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 06/08/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 6/06/07 - 6/07/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	356	23.2	24.1	5.8	8.6	7.57	8.09	104	60	-
DIEPAMHR + organic matter	353	22.9	24.0	5.1	8.5	7.45	8.05	104	60	-
High EC Control @ 15.55 mS/cm + organic matter	14425	23.0	24.0	6.5	8.2	7.56	7.83	1760	140	-
High EC Control @ 21.37 mS/cm + organic matter	20130	22.9	24.2	6.4	8.1	7.61	7.86	2440	120	-
Napa River at Riverside Blvd Terminus (340)	14510	23.1	24.3	6.5	8.6	7.73	7.83	436	134	0.000
Suisun Bay off Chipps Island (508)	21766	23.1	24.4	5.9	8.7	7.63	7.94	236	74	0.001
Suisun Bay, East of middle point (504)	8585	23.2	24.2	6.4	8.8	7.64	7.80	260	82	0.000
Grizzly Bay at Dolphin (602)	8800	23.2	24.1	6.5	8.8	7.62	7.83	264	84	0.000
Carquinez Strait, West of Benicia army dock (405)	19235	23.4	24.4	6.2	8.7	7.53	7.67	308	98	0.000
Montezuma Slough at Nurse Slough (609)	5225	23.4	24.3	6.3	8.5	7.70	7.77	640	120	0.001
Cache Creek / Ulatis Creek Confluence	215	23.0	24.3	5.6	8.4	7.61	7.98	68	74	0.008
DIEPAMHR + 25 ppb PBO	353	23.1	24.3	6.0	8.4	7.60	8.09	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	348	22.8	24.3	6.0	8.3	7.55	8.00	-	-	-
High EC Control @ 15.55 mS/cm + organic matter + 25 ppb PBO	14410	22.7	24.5	6.3	8.1	7.55	7.81	-	-	-
High EC Control @ 21.37 mS/cm + organic matter + 25 ppb PBO	19680	22.7	24.4	6.5	8.0	7.62	7.83	-	-	-
Napa River at Riverside Blvd Terminus (340) + 25 ppb PBO	14405	22.6	24.3	6.5	8.3	7.73	7.82	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	3839	22.5	24.2	6.4	8.5	7.65	7.93	-	-	-
Suisun Bay, East of middle point (504) + 25 ppb PBO	8565	22.6	24.2	6.3	8.6	7.60	7.87	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	8645	22.5	23.3	6.5	8.5	7.61	7.84	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	19115	22.5	24.1	6.4	8.4	7.61	7.68	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	5200	22.5	24.3	6.5	8.8	7.73	7.82	-	-	-
Cache Creek / Ulatis Creek Confluence + 25 ppb PBO	227	22.6	24.5	6.1	8.3	7.70	8.02	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A79-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 06/21/2007 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 6/20/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	98	2.5	97	2.8	NS
DIEPAMHR + organic matter	100	0.0	98	2.5	NS
Sacramento R. at tip of Grand Island (711)	94	3.2	85	6.4	NS
Middle of Broad Slough, west end	95	2.9	98	2.5	NS
Sacramento R. Deep Water Channel, Light 55	85	3.4	87	2.4	NS
Sacramento R. across from Sherman Lake (704)	98	2.5	95	2.9	NS
Old River at mouth of Holland Cut	88	4.8	98	2.5	NS
San Joaquin R., west of Oulton Point	90	4.1	98	2.5	NS
San Joaquin R., between Hog and Turner Cuts (910)	88	6.3	93	4.8	NS
Old River, western arm at railroad bridge	100	0.0	98	2.3	NS
Field Duplicate: Sacramento R. at tip of Grand Island (711)	79	12.1	-	-	NA

	MSD	PMSD
One-way ANOVA	24.27	24.27
Two-way ANOVA	18.04	18.04

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.075	0.005	0.104	0.006	NS
DIEPAMHR + organic matter	0.115	0.001	0.157	0.010	S (134%)
Sacramento R. at tip of Grand Island (711)	0.088	0.007	0.144	0.011	S (164%)
Middle of Broad Slough, west end	0.118	0.010	0.142	0.009	NS
Sacramento R. Deep Water Channel, Light 55	0.120	0.013	0.140	0.004	NS
Sacramento R. across from Sherman Lake (704)	0.122	0.005	0.148	0.007	NS
Old River at mouth of Holland Cut	0.136	0.008	0.097	0.002	NS
San Joaquin R., west of Oulton Point	0.121	0.010	0.101	0.007	NS
San Joaquin R., between Hog and Turner Cuts (910)	0.138	0.007	0.102	0.006	NS
Old River, western arm at railroad bridge	0.134	0.011	0.131	0.006	NS
Field Duplicate: Sacramento R. at tip of Grand Island (711)	0.134	0.006	-	-	NA

	MSD	PMSD
One-way ANOVA	0.04	35.96
Two-way ANOVA	0.04	35.96

- Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$). Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
- NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable

Table A79-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 6/20/07.

Treatment	Field Chemistry					Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	EC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Sacramento R. at tip of Grand Island (711)	173	162	21.9	7.80	8.1	7.0	0.08	0.002
Middle of Broad Slough, west end	2004	1844	21.0	7.94	8.7	11.2	0.03	0.001
Sacramento R. Deep Water Channel, Light 55	189	178	22.1	7.94	8.3	16.3	0.11	0.004
Sacramento R. across from Sherman Lake (704)	2933	2687	20.8	7.95	8.8	17.4	0.09	0.003
Old River at mouth of Holland Cut	292	287	24.3	8.06	7.8	4.0	0.06	0.003
San Joaquin R., west of Oulton Point	261	247	22.4	8.10	8.5	7.2	0.09	0.005
San Joaquin R., between Hog and Turner Cuts (910)	446	441	24.4	7.79	7.1	7.4	0.15	0.004
Old River, western arm at railroad bridge	273	262	22.9	8.10	8.1	8.4	0.09	0.005
Field Duplicate: Sacramento R. at tip of Grand Island (711)	171	162	22.2	7.92	8.1	7.2	0.06	0.002

Table A79-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 06/21/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 6/20/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	342	22.0	23.9	6.7	8.3	7.46	7.98	108	62	-
DIEPAMHR + organic matter	349	22.0	23.3	5.9	8.3	7.39	7.99	108	62	-
Sacramento R. at tip of Grand Island (711)	178	22.2	23.5	5.8	8.7	7.42	7.93	64	62	0.003
Middle of Broad Slough, west end	1942	22.2	23.2	6.2	8.7	7.32	7.80	300	10	0.001
Sacramento R. Deep Water Channel, Light 55	200	22.2	23.5	5.9	8.6	7.51	7.99	76	66	0.005
Sacramento R. across from Sherman Lake (704)	2838	22.3	23.3	6.3	8.7	7.35	7.75	340	50	0.002
Old River at mouth of Holland Cut	302	22.3	23.5	6.0	8.9	7.52	7.89	80	66	0.002
San Joaquin R., west of Oulton Point	258	22.3	23.3	6.0	8.8	7.46	7.89	80	64	0.003
San Joaquin R., between Hog and Turner Cuts (910)	347	22.1	23.5	6.1	8.5	7.43	7.93	124	80	0.006
Old River, western arm at railroad bridge	369	22.0	23.2	6.0	8.8	7.44	7.85	80	64	0.003
Field Duplicate: Sacramento R. at tip of Grand Island (711)	175	22.1	23.4	6.2	8.8	7.50	7.99	68	74	0.003
DIEPAMHR + 25 ppb PBO	339	22.1	23.0	6.5	8.3	7.42	8.01	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	347	22.2	22.9	6.0	8.3	7.40	8.03	-	-	-
Sacramento R. at tip of Grand Island (711) + 25 ppb PBO	175	22.2	23.1	6.2	8.9	7.51	8.02	-	-	-
Middle of Broad Slough, west end + 25 ppb PBO	1922	22.3	23.4	6.1	8.9	7.34	7.83	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	202	22.3	23.6	6.1	8.7	7.51	8.00	-	-	-
Sacramento R. across from Sherman Lake (704) + 25 ppb PBO	2830	22.1	23.2	6.4	8.5	7.36	7.76	-	-	-
Old River at mouth of Holland Cut + 25 ppb PBO	303	22.1	23.6	6.0	8.9	7.51	7.97	-	-	-
San Joaquin R., west of Oulton Point + 25 ppb PBO	259	22.1	23.3	6.2	8.8	7.49	8.00	-	-	-
San Joaquin R., between Hog and Turner Cuts (910) + 25 ppb PBO	349	22.1	23.5	6.1	8.7	7.43	7.89	-	-	-
Old River, western arm at railroad bridge + 25 ppb PBO	363	22.2	23.4	6.5	8.8	7.59	7.84	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A80-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 06/23/2007 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 6/21/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	92	2.6	86	4.8	NS
DIEPAMHR + organic matter	95	2.9	97	2.8	NS
High EC Control @ 14.85 mS/cm	82	7.4	78	10.3	NS
High EC Control @ 17.85 mS/cm	80	4.1	77	3.5	NS
High EC Control @ 19.30 mS/cm	77	7.4	68	16.0	NS
Napa River at Vallejo Seawall (340)	85	7.7	79	7.1	NS
Suisun Bay at Chipps Island (508)	97	2.8	95	2.9	NS
Suisun Bay at Middle Point (504)	87	10.6	85	11.9	NS
Grizzly Bay at Dolphin (602)	92	4.9	70	5.1	NS
Carquinez Straight at Benicia (405)	85	2.7	85	6.4	NS
Montezuma Slough at Nurse Slough (609)	90	4.1	95	2.8	NS

	MSD	PMSD
One-way ANOVA	28.49	29.99
Two-way ANOVA	39.72	41.81

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.061	0.006	0.058	0.011	NS
DIEPAMHR + organic matter	0.078	0.005	0.088	0.009	NS
High EC Control @ 14.85 mS/cm	0.072	0.010	0.063	0.009	NS
High EC Control @ 17.85 mS/cm	0.065	0.006	0.051	0.006	NS
High EC Control @ 19.30 mS/cm	0.037	0.004	0.054	0.008	NS
Napa River at Vallejo Seawall (340)	0.066	0.003	0.066	0.012	NS
Suisun Bay at Chipps Island (508)	0.109	0.021	0.116	0.011	NS
Suisun Bay at Middle Point (504)	0.084	0.010	0.043	0.009	NS
Grizzly Bay at Dolphin (602)	0.057	0.010	0.084	0.004	NS
Carquinez Straight at Benicia (405)	0.052	0.007	0.049	0.007	NS
Montezuma Slough at Nurse Slough (609)	0.105	0.008	0.111	0.008	NS

	MSD	PMSD
One-way ANOVA	0.05	58.24
Two-way ANOVA	0.05	65.63

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$). Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable
3. This high conductivity sample was compared to the High EC control.

Table A80-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 6/21/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Napa River at Vallejo Seawall (340)	17020	25.2	7.76	8.7	5.5	0.02	0.000
Suisun Bay at Chipps Island (508)	5360	21.0	7.97	8.8	24.0	0.10	0.003
Suisun Bay at Middle Point (504)	9520	20.4	8.01	8.8	7.6	0.07	0.002
Grizzly Bay at Dolphin (602)	14090	19.0	7.97	8.9	39.0	0.13	0.003
Carquinez Straight at Benicia (405)	17680	19.0	7.93	8.8	10.1	0.09	0.002
Montezuma Slough at Nurse Slough (609)	7000	20.8	7.84	7.9	23.1	0.12	0.003

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Table A80-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 06/23/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 6/21/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	348.2	23.5	23.6	6.2	8.3	7.59	8.22	108	62	-
DIEPAMHR + organic matter	354.3	23.4	23.9	6.3	8.4	7.58	8.09	108	62	-
High EC Control @ 14.85 mS/cm	14085	23.4	23.7	6.7	8.3	7.46	7.78	1280	78	-
High EC Control @ 17.85 mS/cm	17185	23.5	24.0	6.8	8.5	7.49	7.86	1500	82	-
High EC Control @ 19.30 mS/cm	18495	23.6	23.9	6.9	8.3	7.53	7.86	1820	86	-
Napa River at Vallejo Seawall (340)	16780	23.5	24.1	6.8	8.2	7.75	7.90	2000	141	0.000
Suisun Bay at Chipps Island (508)	5900	23.6	23.9	6.8	8.6	7.62	7.85	500	76	0.003
Suisun Bay at Middle Point (504)	9305	23.6	24.1	6.3	8.5	7.59	7.73	980	82	0.001
Grizzly Bay at Dolphin (602)	13675	23.7	24.1	6.8	8.1	7.62	7.74	3240	120	0.002
Carquinez Straight at Benicia (405)	17820	23.6	24.0	6.6	8.0	7.57	7.70	2000	100	0.002
Montezuma Slough at Nurse Slough (609)	7140	23.5	24.1	6.8	8.9	7.64	7.80	820	70	0.002
DIEPAMHR + 25 ppb PBO	580.05	23.5	23.8	6.5	8.3	7.89	8.16	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	352.45	23.8	24.0	6.7	8.5	7.72	8.03	-	-	-
High EC Control @ 14.85 mS/cm + 25 ppb PBO	14115	23.6	24.0	6.9	8.6	7.51	7.82	-	-	-
High EC Control @ 17.85 mS/cm + 25 ppb PBO	16560	23.7	24.0	6.1	8.3	7.52	7.85	-	-	-
High EC Control @ 19.30 mS/cm + 25 ppb PBO	18705	23.8	23.9	6.7	8.3	7.57	7.82	-	-	-
Napa River at Vallejo Seawall (340) + 25 ppb PBO	16435	24.0	24.1	6.9	8.3	7.79	7.92	-	-	-
Suisun Bay at Chipps Island (508) + 25 ppb PBO	5905	23.8	24.0	6.7	8.3	7.67	7.84	-	-	-
Suisun Bay at Middle Point (504) + 25 ppb PBO	9310	23.8	24.0	7.1	8.4	7.59	7.81	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	13915	23.4	24.2	7.0	8.3	7.64	7.76	-	-	-
Carquinez Straight at Benicia (405) + 25 ppb PBO	17885	23.3	24.2	6.9	8.0	7.62	7.72	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	7125	23.7	24.3	7.0	8.5	7.69	7.87	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A81-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 07/12/07 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 7/10/07.

Unmanipulated Samples

Sample	Mean Survival (%) ¹					Weight (mg/surviving individual) ¹	
	24 hr	48 hr	72 hr	96 hr	10 day	mean	se
DIEPAMHR	100	100	100	100	100	0.056	0.002
DIEPAMHR + organic matter	98	95	95	95	95	0.053	0.005
High EC Control @ 15.4 mS/cm	98	93	90	85	50	0.067	0.025
High EC Control @ 22.75 mS/cm	88	58	33	30	18	0.056	0.010
Middle of Broad Slough, West end (804)	98	93	90	88	88	0.069	0.007
Montezuma Slough @ Nurse Slough (609)	98	90	77	77	75	0.063	0.006
Grizzly Bay @ Dolphin (602) ²	71	49	36	33	25	0.073	0.025
Carquinez Strait, West of Benicia army dock (405) ³	5	3	0	0	0	-	-
Suisun Bay, East of middle point (504)	93	93	90	90	88	0.067	0.009
Suisun Bay, off Chipps Island (508)	100	100	98	93	93	0.065	0.005
Sacramento R., across from Sherman Lake (704)	98	98	98	98	98	0.070	0.007

PBO-added treatments

Sample	Mean Survival (%) ¹				
	24 hr	48 hr	72 hr	96 hr	10 day
DIEPAMHR	78	30	0	0	-
DIEPAMHR + organic matter	69	39	3	0	-
High EC Control @ 15.4 mS/cm	74	29	3	0	-
High EC Control @ 22.75 mS/cm	31	10	0	0	-
Middle of Broad Slough, West end (804)	43	0	0	0	-
Montezuma Slough @ Nurse Slough (609)	93	83	70	-	-
Grizzly Bay @ Dolphin (602) ²	48	18	5	-	-
Carquinez Strait, West of Benicia army dock (405) ³	0	0	0	-	-
Suisun Bay, East of middle point (504)	60	5	0	-	-
Suisun Bay, off Chipps Island (508)	80	8	0	-	-
Sacramento R., across from Sherman Lake (704)	90	65	15	-	-

1. Highlighted cells indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. This high conductivity sample was compared to the 15.4 mS/cm High EC control.

3. This high conductivity sample was compared to the 22.75 mS/cm High EC control.

Table A81-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 7/10/07.

Treatment	Field Chemistry				Un-ionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)	
Middle of Broad Slough, West end (804)	898	21.6	8.13	8.4	0.001
Montezuma Slough @ Nurse Slough (609)	7660	-	7.85	7.9	0.001
Grizzly Bay @ Dolphin (602)	15030	20.0	7.96	8.6	0.003
Carquinez Strait, West of Benicia army dock (405)	21930	20.0	7.95	8.3	0.003
Suisun Bay, East of middle point (504)	7150	21.0	8.03	8.6	0.003
Suisun Bay, off Chipps Island (508)	4776	21.0	7.96	8.5	0.002
Sacramento R., across from Sherman Lake (704)	480	21.1	7.97	8.7	0.004

Table A81-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 07/12/2007 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 7/10/07.

Treatment	Laboratory Chemistry						Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH			Max pH
DIEPAMHR	337	23.8	24.6	6.8	8.0	7.48	8.18	104	60
DIEPAMHR + organic matter	340	23.0	24.6	6.7	8.5	7.41	8.12	-	-
High EC Control @ 15.4 mS/cm	14275	24.0	24.5	6.4	8.5	7.38	7.84	860	78
High EC Control @ 22.75 mS/cm	21070	23.0	24.6	6.6	8.6	7.48	7.89	1300	85
Middle of Broad Slough, West end (804)	934	24.1	24.6	6.8	8.6	7.51	8.00	120	60
Montezuma Slough @ Nurse Slough (609)	7125	24.3	24.6	6.9	8.8	7.54	7.76	180	60
Grizzly Bay @ Dolphin (602)	12935	23.9	24.6	6.6	8.0	7.41	7.75	1640	100
Carquinez Strait, West of Benicia army dock (405)	19135	22.7	24.2	7.0	7.7	7.40	7.80	2480	80
Suisun Bay, East of middle point (504)	6770	24.1	24.7	6.9	8.9	7.57	7.97	760	50
Suisun Bay, off Chipps Island (508)	4598	24.1	24.7	6.8	8.6	7.52	7.83	440	70
Sacramento R., across from Sherman Lake (704)	557	24.0	24.7	6.5	8.4	7.50	8.02	112	52
DIEPAMHR + 25 ppb PBO	347	22.7	23.0	6.7	8.4	7.51	8.28	-	-
DIEPAMHR + organic matter + 25 ppb PBO	338	22.6	23.2	7.0	8.4	7.62	8.21	-	-
High EC Control @ 15.4 mS/cm + 25 ppb PBO	14235	22.7	23.4	6.6	8.2	7.34	7.94	-	-
High EC Control @ 22.75 mS/cm + 25 ppb PBO	21315	23.0	23.5	7.0	7.7	7.43	7.92	-	-
Middle of Broad Slough, West end (804) + 25 ppb PBO	4866	23.5	23.7	7.5	8.6	7.66	8.12	-	-
Montezuma Slough @ Nurse Slough (609) + 25 ppb PBO	7370	23.7	23.7	8.1	8.1	7.98	7.98	-	-
Grizzly Bay @ Dolphin (602) + 25 ppb PBO	13655	23.1	23.8	6.7	7.9	7.39	7.80	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	21010	23.2	24.1	7.0	7.8	7.48	7.81	-	-
Suisun Bay, East of middle point (504) + 25 ppb PBO	6795	22.9	24.0	6.9	8.1	7.51	8.02	-	-
Suisun Bay, off Chipps Island (508) + 25 ppb PBO	4543	23.4	24.1	6.7	8.2	7.49	7.98	-	-
Sacramento R., across from Sherman Lake (704) + 25 ppb PBO	480	24.1	24.1	8.3	8.3	8.05	8.05	-	-

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Table A82-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 07/13/2007 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 7/11/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	95	3.1	98	2.5	NS
DIEPAMHR + organic matter	97	2.8	95	5.0	NS
Sacramento R. Deep Water Channel, Light 55	98	2.5	98	2.5	NS
Old River at Holland Cut (915)	98	2.5	98	2.5	NS
San Joaquin R. West of Oulton Point (812)	95	2.9	98	2.5	NS
San Joaquin R. between Hog and Turner Cuts (910)	95	2.9	95	2.9	NS
Old River, Western arm at railroad bridge (902)	100	0.0	100	0.0	NS
Sacramento R. at tip of Grand Island (711)	95	2.9	98	2.5	NS

	MSD	PMSD
One-way ANOVA	12.23	12.58
Two-way ANOVA	14.00	14.56

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.044	0.001	0.049	0.002	NS
DIEPAMHR + organic matter	0.069	0.003	0.070	0.007	NS
Sacramento R. Deep Water Channel, Light 55	0.098	0.015	0.098	0.008	NS
Old River at Holland Cut (915)	0.107	0.009	0.102	0.004	NS
San Joaquin R. West of Oulton Point (812)	0.094	0.006	0.090	0.009	NS
San Joaquin R. between Hog and Turner Cuts (910)	0.107	0.005	0.093	0.005	NS
Old River, Western arm at railroad bridge (902)	0.093	0.002	0.096	0.008	NS
Sacramento R. at tip of Grand Island (711)	0.078	0.006	0.074	0.008	NS

	MSD	PMSD
One-way ANOVA	0.03	49.47
Two-way ANOVA	0.04	51.40

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable

Table A82-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 7/11/07.

Treatment	Field Chemistry				Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)		
Sacramento R. Deep Water Channel, Light 55	249	22.0	8.29	8.2	0.070	0.005
Old River at Holland Cut (915)	246	22.0	8.21	7.6	0.060	0.004
San Joaquin R. West of Oulton Point (812)	242	21.8	8.18	8.4	0.070	0.004
San Joaquin R. between Hog and Turner Cuts (910)	284	23.6	8.00	6.8	0.060	0.003
Old River, Western arm at railroad bridge (902)	352	20.8	8.26	7.9	0.080	0.005
Sacramento R. at tip of Grand Island (711)	126	21.5	8.03	8.2	0.180	0.008

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Table A82-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 7/13/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 7/11/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	348	23.6	24.6	6.1	8.4	7.42	7.87	104	60	-
DIEPAMHR + organic matter	350	23.5	24.4	6.7	8.5	7.44	7.94	104	60	-
Sacramento R. Deep Water Channel, Light 55	257	23.8	24.6	6.2	8.5	7.48	7.86	78	75	0.002
Old River at Holland Cut (915)	260	23.7	24.4	6.3	8.8	7.51	7.76	62	62	0.002
San Joaquin R. West of Oulton Point (812)	223	23.6	24.4	6.3	8.6	7.36	7.74	58	56	0.002
San Joaquin R. between Hog and Turner Cuts (910)	298	23.4	24.6	6.2	8.4	7.39	7.67	80	67	0.001
Old River, Western arm at railroad bridge (902)	299	23.8	24.4	6.4	8.4	7.40	7.66	66	59	0.002
Sacramento R. at tip of Grand Island (711)	139	23.5	24.6	5.9	8.4	7.38	7.61	52	55	0.003
DIEPAMHR + 25 ppb PBO	349	22.9	24.4	6.6	8.3	7.43	7.97	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	345	23.0	24.4	6.4	8.3	7.42	7.93	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	255	23.0	24.4	6.6	8.9	7.53	7.99	-	-	-
Old River at Holland Cut (915) + 25 ppb PBO	257	22.8	24.7	6.6	8.8	7.45	7.85	-	-	-
San Joaquin R. West of Oulton Point (812) + 25 ppb PBO	219	22.9	24.4	6.2	8.9	7.36	7.81	-	-	-
San Joaquin R. between Hog and Turner Cuts (910) + 25 ppb PBO	295	22.8	24.3	6.3	8.7	7.40	7.70	-	-	-
Old River, Western arm at railroad bridge (902) + 25 ppb PBO	294	22.9	24.4	6.4	8.5	7.36	7.76	-	-	-
Sacramento R. at tip of Grand Island (711) + 25 ppb PBO	141	22.8	24.4	6.2	8.5	7.46	7.74	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A83-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 07/26/07 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 7/25/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	98	2.5	100	0.0	NS
DIEPAMHR + organic matter	95	2.9	95	2.9	NS
High EC Control @ 14.5 mS/cm	72	7.3	54	8.2	NS
High EC Control @ 21.81 mS/cm	49	7.6	25	12.6	NS
Montezuma Slough at Nurse Slough (609)	70	23.5	100	0.0	NS
Middle of Broad Slough, West end (804)	100	0.0	100	0.0	NS
Sacramento R. across from Sherman Lake (704)	95	2.9	100	0.0	NS
Suisun Bay, East of middle point (504)	90	4.1	95	2.9	NS
Grizzly Bay at Dolphin (602) ³	95	5.0	90	7.1	NS
Carquinez Strait, West of Benicia army dock (405) ⁴	59	7.2	33	14.9	NS
Suisun Bay off Chipps Island (508)	93	2.4	100	0.0	NS

	MSD	PMSD
One-way ANOVA	41.3	43.5
Two-way ANOVA	27.6	29.1

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.040	0.007	0.056	0.002	NS
DIEPAMHR + organic matter	0.080	0.003	0.063	0.006	NS
High EC Control @ 14.5 mS/cm	0.047	0.007	0.046	0.004	NS
High EC Control @ 21.81 mS/cm	0.027	0.002	0.023	0.010	NS
Montezuma Slough at Nurse Slough (609)	0.052	0.001	0.077	0.005	NS
Middle of Broad Slough, West end (804)	0.056	0.013	0.087	0.008	NS
Sacramento R. across from Sherman Lake (704)	0.086	0.004	0.079	0.005	NS
Suisun Bay, East of middle point (504)	0.054	0.003	0.058	0.013	NS
Grizzly Bay at Dolphin (602) ³	0.055	0.005	0.058	0.011	NS
Carquinez Strait, West of Benicia army dock (405) ⁴	0.039	0.005	0.039	0.007	NS
Suisun Bay off Chipps Island (508)	0.051	0.005	0.050	0.006	NS

	MSD	PMSD
One-way ANOVA	0.029	36.1
Two-way ANOVA	0.036	44.5

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable

3. This high conductivity sample was compared to the High EC control @ 14.5 mS/cm.

4. This high conductivity sample was compared to the High EC control @ 21.81 mS/cm.

Table A83-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 7/25/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Montezuma Slough at Nurse Slough (609)	8150	22.9	7.73	8.2	12.7	0.010	0.000
Middle of Broad Slough, West end (804)	1087	22.0	8.01	8.3	8.3	0.030	0.001
Sacramento R. across from Sherman Lake (704)	159	22.0	7.96	8.7	9.6	0.040	0.002
Suisun Bay, East of middle point (504)	9270	22.6	7.85	8.5	12.7	0.000	0.000
Grizzly Bay at Dolphin (602)	14230	22.3	7.83	8.3	15.9	0.010	0.000
Carquinez Strait, West of Benicia army dock (405)	21500	21.5	7.81	8.2	7.5	0.030	0.001
Suisun Bay off Chipps Island (508)	5030	22.6	7.89	8.4	9.0	0.010	0.000

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Table A83-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 7/26/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 7/25/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	342.3	23.6	23.8	6.4	8.1	7.59	8.06	108	59	-
DIEPAMHR + organic matter	345	23.7	23.9	6.5	8.3	7.52	8.04	108	59	-
High EC Control @ 14.5 mS/cm	13260	23.7	23.9	6.4	8.3	7.44	7.76	-	-	-
High EC Control @ 21.81 mS/cm	20275	23.7	23.9	6.5	8.2	7.55	7.75	-	-	-
Montezuma Slough at Nurse Slough (609)	7605	23.4	23.8	6.6	8.4	7.62	7.77	760	95	0.000
Middle of Broad Slough, West end (804)	1067	23.8	23.8	6.5	8.9	7.52	7.78	144	60	0.001
Sacramento R. across from Sherman Lake (704)	201.25	23.7	23.9	6.2	8.6	7.48	7.98	56	56	0.002
Suisun Bay, East of middle point (504)	8905	23.8	23.9	6.5	8.2	7.47	7.77	944	74	0.000
Grizzly Bay at Dolphin (602)	13550	23.8	23.8	6.6	8.2	7.48	7.7	1380	82	0.000
Carquinez Strait, West of Benicia army dock (405)	19830	23.8	23.8	6.2	8.5	7.51	7.68	400	94	0.000
Suisun Bay off Chipps Island (508)	4845	23.7	23.9	6.6	8.3	7.47	7.84	532	66	0.000
DIEPAMHR + 25 ppb PBO	463.05	23.3	23.9	6.5	8.2	7.58	8.17	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	353.5	23.1	23.9	6.3	8.3	7.51	8.04	-	-	-
High EC Control @ 14.5 mS/cm + 25 ppb PBO	13215	23.2	23.9	6.5	8.1	7.48	7.81	-	-	-
High EC Control @ 21.81 mS/cm + 25 ppb	20000	23.4	23.9	6.6	7.8	7.58	7.81	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	7510	23.2	23.8	6.2	8.2	7.58	7.86	-	-	-
Middle of Broad Slough, West end (804) + 25 ppb PBO	4451.5	23.2	23.8	6.4	8.5	7.53	7.84	-	-	-
Sacramento R. across from Sherman Lake (704) + 25 ppb PBO	215.45	23.7	24.2	6.2	8.8	7.5	7.99	-	-	-
Suisun Bay, East of middle point (504) + 25 ppb PBO	8765	23.9	24.2	6.3	8.6	7.43	7.63	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	13515	23.8	24.2	6.3	8.8	7.52	7.66	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	20030	23.8	24.2	6.2	8.5	7.54	7.72	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	4911.5	23.8	24.5	6.5	8.9	7.49	7.78	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A84-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 07/27/2007 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 7/25/07 - 7/26/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO ²
DIEPAMHR	100	0.0	100	0.0	NS
DIEPAMHR + organic matter	98	2.3	100	0.0	NS
High EC Control @ 24.27 mS/cm	83	7.5	77	7.3	NS
Napa River at Riverside Blvd Terminus (340) ³	67	10.4	44	8.7	S
San Joaquin R., West of Oulton Point (812)	100	0.0	100	0.0	NS
San Joaquin R. between Hog and Turner Cuts (910)	100	0.0	100	0.0	NS
Old River at mouth of Holland Cut (915)	100	0.0	100	0.0	NS
Old River, western arm at railroad bridge (902)	98	2.5	100	0.0	NS
Sacramento R. Deep Water Channel, Light 55	92	5.3	100	0.0	NS
Sacramento R. at tip of Grand Island (711)	98	2.5	95	2.9	NS
Field Dup: Old River, western arm at railroad bridge (902)	100	0.0	-	-	NA

	MSD	PMSD
One-way ANOVA	21.4	21.9
Two-way ANOVA	21.8	22.3

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO ²
DIEPAMHR	0.063	0.004	0.074	0.007	NS
DIEPAMHR + organic matter	0.086	0.013	0.108	0.007	NS
High EC Control @ 24.27 mS/cm	0.073	0.005	0.077	0.015	NS
Napa River at Riverside Blvd Terminus (340) ³	0.043	0.015	0.084	0.024	NS
San Joaquin R., West of Oulton Point (812)	0.124	0.012	0.118	0.014	NS
San Joaquin R. between Hog and Turner Cuts (910)	0.129	0.008	0.113	0.012	NS
Old River at mouth of Holland Cut (915)	0.136	0.014	0.135	0.009	NS
Old River, western arm at railroad bridge (902)	0.118	0.011	0.128	0.017	NS
Sacramento R. Deep Water Channel, Light 55	0.134	0.016	0.132	0.013	NS
Sacramento R. at tip of Grand Island (711)	0.153	0.005	0.129	0.008	NS
Field Dup: Old River, western arm at railroad bridge (902)	0.117	0.007	-	-	NA

	MSD	PMSD
One-way ANOVA	0.053	62.1
Two-way ANOVA	0.066	76.6

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.
Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable
3. This high conductivity sample was compared to the High EC control @ 24.27 mS/cm.

Table A84-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 7/25//07- 7/26/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Napa River at Riverside Blvd Terminus (340)	24400	23.8	7.64	6.6	20.6	0.030	0.000
San Joaquin R., West of Oulton Point (812)	281	22.4	7.21	8.4	5.7	0.080	0.021
San Joaquin R. between Hog and Turner Cuts (910)	232	23.8	7.41	7.3	7.5	0.060	0.001
Old River at mouth of Holland Cut (915)	328	23.8	7.39	8.0	5.9	0.040	0.000
Old River, western arm at railroad bridge (902)	397	23.9	7.56	8.4	7.5	0.020	0.000
Sacramento R. Deep Water Channel, Light 55	263	23.1	7.32	8.5	42.2	0.060	0.001
Sacramento R. at tip of Grand Island (711)	142	22.6	7.29	8.1	11.1	0.220	0.002
Field Dup: Old River, western arm at railroad bridge (902)	397	23.9	7.58	8.4	5.8	0.030	0.001

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Table A84-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 07/27/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 7/25/07 - 7/26/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	348.55	21.5	23.1	6.5	8.3	7.51	8.12	108	59	-
DIEPAMHR + organic matter	346.95	21.5	23.7	6.8	8.3	7.45	8.09	108	59	-
High EC Control @ 24.27 mS/cm	22380	21.5	23.5	6.9	8.2	7.54	7.74	-	-	-
Napa River at Riverside Blvd Terminus (340)	22800	21.5	23.9	6.7	8.0	7.42	7.84	3080	139	0.000
San Joaquin R., West of Oulton Point (812)	273.8	21.4	23.8	6.3	8.8	7.56	7.67	68	57	0.002
San Joaquin R. between Hog and Turner Cuts (910)	237.6	21.4	23.9	6.6	8.5	7.54	7.74	68	63	0.001
Old River at mouth of Holland Cut (915)	330.1	21.4	23.9	6.8	8.5	7.51	7.68	72	59	0.001
Old River, western arm at railroad bridge (902)	384.15	21.4	24.2	6.6	8.7	7.48	7.90	76	58	0.001
Sacramento R. Deep Water Channel, Light 55	207.5	21.4	24.3	6.6	8.5	7.60	7.92	80	75	0.002
Sacramento R. at tip of Grand Island (711)	194.25	21.4	24.2	6.5	8.5	7.47	7.69	52	57	0.003
Field Dup: Old River, western arm at railroad bridge (902)	384.25	21.4	24.2	6.7	8.6	7.47	7.86	68	58	0.001
DIEPAMHR + 25 ppb PBO	327.55	21.4	23.2	6.6	8.2	7.50	8.07	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	331.4	21.4	23.5	6.6	8.4	7.46	8.00	-	-	-
High EC Control @ 24.27 mS/cm + 25 ppb PBO	22050	21.4	23.4	6.7	7.9	7.50	7.74	-	-	-
Napa River at Riverside Blvd Terminus (340) + 25 ppb PBO	22305	21.3	23.3	6.5	8.2	7.45	7.84	-	-	-
San Joaquin R., West of Oulton Point (812) + 25 ppb PBO	276.35	21.3	23.1	6.6	8.6	7.52	7.77	-	-	-
San Joaquin R. between Hog and Turner Cuts (910) + 25 ppb PBO	233	21.3	23.1	6.3	8.5	7.50	7.76	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	330.4	21.2	23.0	6.6	8.4	7.47	7.70	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	373.1	21.0	22.8	6.5	8.5	7.46	7.86	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	219.35	21.0	22.8	6.6	8.7	7.60	7.98	-	-	-
Sacramento R. at tip of Grand Island (711) + 25 ppb PBO	195.85	21.4	23.1	6.4	8.7	7.45	7.74	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A85-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 08/09/2007 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 8/08/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	98	2.5	98	2.5	NS
DIEPAMHR + organic matter	93	4.8	98	2.5	NS
High EC Control @ 11.68 mS/cm	100	0.0	95	3.1	NS
High EC Control @ 15.17 mS/cm	98	2.5	85	6.4	NS
High EC Control @ 23.9 mS/cm	31	6.6	5	5.0	S
Montezuma Slough at Nurse Slough (609)	95	2.9	100	0.0	NS
Middle of Broad Slough, West end (804)	98	2.5	100	0.0	NS
Grizzly Bay at Dolphin (602) ³	87	7.7	100	0.0	NS
Carquinez Strait, West of Benicia army dock (405) ⁴	56	2.4	29	3.2	S
Suisun Bay off Chipps Island (508)	100	0.0	98	2.5	NS
Suisun Bay, East of middle point (504) ⁵	95	5.0	92	8.3	NS
Field Duplicate: Suisun Bay, East of middle point (504) ⁵	95	5.0	-	-	NA

	MSD	PMSD
One-way ANOVA	20.8	22.5
Two-way ANOVA	21.4	23.1

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.060	0.004	0.060	0.005	NS
DIEPAMHR + organic matter	0.092	0.005	0.074	0.007	NS
High EC Control @ 11.68 mS/cm	0.065	0.003	0.070	0.005	NS
High EC Control @ 15.17 mS/cm	0.058	0.006	0.048	0.002	NS
High EC Control @ 23.9 mS/cm	0.049	0.002	0.075	-	NA
Montezuma Slough at Nurse Slough (609)	0.087	0.007	0.073	0.008	NS
Middle of Broad Slough, West end (804)	0.082	0.006	0.061	0.010	NS
Grizzly Bay at Dolphin (602) ³	0.058	0.005	0.058	0.005	NS
Carquinez Strait, West of Benicia army dock (405) ⁴	0.046	0.004	0.052	0.007	NS
Suisun Bay off Chipps Island (508)	0.063	0.007	0.064	0.007	NS
Suisun Bay, East of middle point (504) ⁵	0.064	0.005	0.061	0.004	NS
Field Duplicate: Suisun Bay, East of middle point (504) ⁵	0.053	0.002	-	-	NA

	MSD	PMSD
One-way ANOVA	0.025	30.0
Two-way ANOVA	0.031	33.2

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.
Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable
3. This high conductivity sample was compared to the 15.17 mS/cm High EC control.
4. This high conductivity sample was compared to the 23.9 mS/cm High EC control.
5. This high conductivity sample was compared to the 11.68 mS/cm High EC control.

Table A85-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 8/08/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Montezuma Slough at Nurse Slough (609)	8970	20.3	7.43	8.3	31.1	0.030	0.000
Middle of Broad Slough, West end (804)	1802	20.7	7.65	8.3	10.5	0.020	0.000
Grizzly Bay at Dolphin (602)	14440	19.4	7.69	8.8	25.2	0.080	0.001
Carquinez Strait, West of Benicia army dock (405)	22310	19.4	7.81	8.5	9.1	0.110	0.002
Suisun Bay off Chipps Island (508)	7140	20.8	7.88	8.7	19.3	0.070	0.002
Suisun Bay, East of middle point (504)	9980	20.5	7.86	8.8	8.6	0.040	0.001
Field Duplicate: Suisun Bay, East of middle point (504)	9980	20.8	7.86	8.8	8.9	0.060	0.001

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Table A85-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 8/09/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 8/08/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	351	23.4	23.8	6.5	8.4	7.63	8.19	104	57	-
DIEPAMHR + 1% Delta Water Concentrate	347	23.8	23.8	6.2	8.3	7.59	8.21	104	57	-
High EC Control @ 11.68 mS/cm	11300	23.7	23.9	7.0	8.4	7.49	8.01	1360	100	-
High EC Control @ 15.17 mS/cm	14445	23.7	23.8	7.0	8.1	7.55	8.01	1880	120	-
High EC Control @ 23.9 mS/cm	22780	23.8	23.8	6.5	8.1	7.54	8.05	2960	120	-
Montezuma Slough at Nurse Slough (609)	8690	23.8	23.8	6.3	8.3	7.62	7.95	1020	110	0.001
Middle of Broad Slough, West end (804)	1774	23.7	23.8	6.3	8.8	7.57	8.11	80	80	0.001
Grizzly Bay at Dolphin (602)	13945	23.7	23.9	6.5	8.3	7.54	7.87	1760	120	0.002
Carquinez Strait, West of Benicia army dock (405)	21625	23.7	23.9	6.4	8.6	7.52	7.86	2720	140	0.003
Suisun Bay off Chipps Island (508)	3835	23.8	23.8	6.6	8.6	7.51	7.94	152	70	0.003
Suisun Bay, East of middle point (504)	10185	23.7	23.8	6.4	8.5	7.53	7.91	1080	120	0.001
Field Duplicate: Suisun Bay, East of middle point (504)	10500	23.7	23.7	6.3	8.9	7.54	7.87	1240	120	0.002
DIEPAMHR + 25 ppb PBO	354	22.9	23.7	6.7	8.3	7.66	8.21	-	-	-
DIEPAMHR + 1% Delta Water Concentrate + 25 ppb PBO	16284	23.1	23.7	6.6	8.4	7.63	8.22	-	-	-
High EC Control @ 11.68 mS/cm + 25 ppb PBO	10835	23.1	23.7	7.0	8.3	7.55	8.07	-	-	-
High EC Control @ 15.17 mS/cm + 25 ppb PBO	14290	22.7	23.7	7.0	8.4	7.53	8.01	-	-	-
High EC Control @ 23.9 mS/cm + 25 ppb PBO	21790	23.1	23.8	6.9	8.0	7.57	8.00	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	15320	23.0	23.9	6.7	8.7	7.69	7.87	-	-	-
Middle of Broad Slough, West end (804) + 25 ppb PBO	5319	23.1	23.9	6.7	8.9	7.59	8.08	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	7670	23.1	24.1	6.3	8.7	7.53	7.89	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	17625	23.0	24.0	6.4	8.3	7.52	7.83	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	6755	23.0	24.0	6.3	8.4	7.59	7.96	-	-	-
Suisun Bay, East of middle point (504) + 25 ppb PBO	10190	23.0	24.0	6.7	8.6	7.54	8.89	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A86-1. Survival of *H. azteca* in a Toxicity Identification Evaluation initiated on 8/16/07 examining the cause of toxicity in an ambient water column sample collected at POD site 405 (the Carquinez Strait, West of the Benicia army dock) by the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 8/8/07.¹

Treatment	% Survival ²									
	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Day 8	Day 9	Day 10
DIEPAMHR	100	100	100	100	100	100	100	100	100	100
DIEPAMHR @ 15°C	100	100	100	100	100	97	97	97	93	93
High EC DIEPAMHR @ 21.16 mS	100	88	80	80	73	65	50	47	31	28
High EC DIEPAMHR @ 15°C	100	93	93	80	57	47	37	30	27	20
High EC DIEPAMHR + MeOH @ 0.5%	100	96	90	90	90	86	76	65	41	34
High EC DIEPAMHR + eluate addback @ 3X	100	73	65	51	41	41	41	35	14	10
High EC DIEPAMHR + 50 ppb PBO	100	96	82	82	71	68	61	54	30	30
High EC DIEPAMHR + 500X esterase	100	92	92	92	92	92	92	92	92	92
High EC DIEPAMHR + 500X BSA	95	72	39	29	29	29	29	24	24	19
DIEPAMHR C8 Blank	97	97	97	90	90	90	90	90	90	90
POD 405	97	97	87	87	83	83	77	77	77	77
POD 405 @ 15°C	97	97	97	83	70	57	57	50	50	47
POD 405 + 50 ppb PBO	96	79	63	63	58	58	58	58	58	58
POD 405 + 500X esterse	83	83	80	80	77	73	73	73	73	73
POD 405 + 500X BSA	100	47	43	27	23	19	19	8	8	0
Sample C8 Rinsate	97	87	87	73	73	73	73	73	73	73

1) Sample was treated through Varian C8 column on 08/14/07.

2) Highlighted cells indicate less than 50% survival of test organisms.

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Table A87-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 08/10/2007 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 8/09/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO ²
DIEPAMHR	100	0.0	98	2.5	NS
DIEPAMHR + organic matter	98	2.5	93	4.8	NS
San Joaquin R., West of Oulton Point (812)	95	2.9	90	4.1	NS
Old River at mouth of Holland Cut (915)	98	2.5	95	2.9	NS
San Joaquin R., between Hog and Turner Cuts (910)	100	0.0	98	2.5	NS
Old River, western arm at railroad bridge (902)	92	2.6	93	2.5	NS
Sacramento R. at tip of Grand Island (711)	90	7.1	90	5.8	NS
Sacramento R. across from Sherman Lake (704)	98	2.5	98	2.5	NS
Sacramento R. Deep Water Channel, Light 55	95	2.9	98	2.5	NS
Field Duplicate: Old R. at mouth of Holland Cut (915)	100	0.0	-	-	NA

	MSD	PMSD
One-way ANOVA	14.6	15.0
Two-way ANOVA	17.5	18.0

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO ²
DIEPAMHR	0.076	0.006	0.068	0.007	NS
DIEPAMHR + organic matter	0.081	0.005	0.102	0.013	NS
San Joaquin R., West of Oulton Point (812)	0.111	0.005	0.084	0.005	NS
Old River at mouth of Holland Cut (915)	0.102	0.006	0.106	0.016	NS
San Joaquin R., between Hog and Turner Cuts (910)	0.107	0.014	0.115	0.012	NS
Old River, western arm at railroad bridge (902)	0.092	0.011	0.090	0.008	NS
Sacramento R. at tip of Grand Island (711)	0.099	0.009	0.088	0.010	NS
Sacramento R. across from Sherman Lake (704)	0.101	0.009	0.101	0.008	NS
Sacramento R. Deep Water Channel, Light 55	0.119	0.034	0.095	0.010	NS
Field Duplicate: Old R. at mouth of Holland Cut (915)	0.100	0.009	-	-	NA

	MSD	PMSD
One-way ANOVA	0.064	79.8
Two-way ANOVA	0.063	78.3

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.
Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable

Table A87-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 8/09/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
San Joaquin R., West of Oulton Point (812)	360	21.9	7.86	8.5	3.0	0.080	0.002
Old River at mouth of Holland Cut (915)	397	22.8	7.65	8.2	2.8	0.050	0.001
San Joaquin R., between Hog and Turner Cuts (910)	325	24.3	7.54	7.0	4.2	0.050	0.001
Old River, western arm at railroad bridge (902)	576	22.6	7.81	8.9	3.0	0.040	0.001
Sacramento R. at tip of Grand Island (711)	143	22.1	7.86	8.6	2.3	0.200	0.006
Sacramento R. across from Sherman Lake (704)	1603	21.4	7.73	8.7	8.6	0.080	0.002
Sacramento R. Deep Water Channel, Light 55	237	22.3	7.96	8.6	17.5	0.080	0.003
Field Duplicate: Old R. at mouth of Holland Cut (915)	397	22.8	7.65	8.2	2.9	0.040	0.001

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Table A87-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 08/10/2007 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 8/09/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	350	23.7	23.9	6.3	8.1	7.50	8.13	104	60	-
DIEPAMHR + organic matter	346	23.4	23.8	6.5	8.3	7.49	8.14	104	60	-
San Joaquin R., West of Oulton Point (812)	363	23.8	24.0	6.3	8.6	7.48	7.98	68.0	58.0	0.004
Old River at mouth of Holland Cut (915)	376	23.8	23.8	6.7	8.2	7.53	7.99	108	58	0.002
San Joaquin R., between Hog and Turner Cuts (910)	331	23.8	23.8	6.4	8.8	7.57	7.70	44	70	0.001
Old River, western arm at railroad bridge (902)	574	23.8	23.8	6.6	8.6	7.51	8.11	92	58	0.002
Sacramento R. at tip of Grand Island (711)	155	23.8	23.8	6.3	8.6	7.51	8.14	52	58	0.013
Sacramento R. across from Sherman Lake (704)	1576	23.8	23.9	6.6	8.7	7.48	7.89	192	70	0.003
Sacramento R. Deep Water Channel, Light 55	239	23.8	23.8	6.4	8.2	7.62	8.15	72	60	0.005
Field Duplicate: Old R. at mouth of Holland Cut (915)	385	23.8	23.8	6.4	8.4	7.51	7.86	76	58	0.001
DIEPAMHR + 25 ppb PBO	346	23.7	23.8	6.7	8.2	7.54	8.13	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	344	23.5	23.8	6.6	8.3	7.50	8.09	-	-	-
San Joaquin R., West of Oulton Point (812) + 25 ppb PBO	360	23.2	23.8	6.6	8.9	7.51	7.97	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	366	23.2	23.8	6.5	8.4	7.54	7.92	-	-	-
San Joaquin R., between Hog and Turner Cuts (910) + 25 ppb PBO	333	23.2	23.9	6.4	8.4	7.59	7.91	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	571	23.2	23.8	6.5	8.6	7.52	8.11	-	-	-
Sacramento R. at tip of Grand Island (711) + 25 ppb PBO	155	23.2	23.9	6.7	8.7	7.62	7.87	-	-	-
Sacramento R. across from Sherman Lake (704) + 25 ppb PBO	1558	23.2	24.0	6.8	8.8	7.51	7.97	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	241	23.1	24.2	6.8	8.4	7.65	8.12	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A88-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 8/23/07 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 8/22/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	100	0.0	100	0.0	NS
DIEPAMHR + organic matter	90	4.1	92	5.3	NS
High EC Control @ 12.53 mS/cm	100	0.0	90	4.1	NS
High EC Control @ 17.15 mS/cm	93	2.5	100	0.0	NS
High EC Control @ 24.14 mS/cm	75	9.6	48	13.4	NS
Montezuma Slough at Nurse Slough (609)	93	2.4	100	0.0	NS
Suisun Bay, East of middle point (504) ³	98	2.5	95	2.9	NS
Grizzly Bay at Dolphin (602) ⁴	83	7.8	82	5.5	NS
Sacramento R. across from Sherman Lake (704)	100	0.0	95	2.9	NS
Middle of Broad Slough, West end (804)	98	2.5	98	2.5	NS
Carquinez Strait, West of Benicia army dock (405) ⁵	29	18.2	28	6.5	NS
Suisun Bay off Chipps Island (508)	95	5.0	97	2.8	NS
Field Duplicate: Middle of Broad Slough, West end (804)	98	2.5	-	-	NA

	MSD	PMSD
One-way ANOVA	32.6	36.9
Two-way ANOVA	32.5	36.1

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.058	0.003	0.043	0.002	NS
DIEPAMHR + organic matter	0.084	0.011	0.078	0.003	NS
High EC Control @ 12.53 mS/cm	0.076	0.004	0.065	0.011	NS
High EC Control @ 17.15 mS/cm	0.065	0.003	0.065	0.004	NS
High EC Control @ 24.14 mS/cm	0.052	0.009	0.055	0.002	NS
Montezuma Slough at Nurse Slough (609)	0.076	0.005	0.067	0.004	NS
Suisun Bay, East of middle point (504) ³	0.067	0.001	0.053	0.010	NS
Grizzly Bay at Dolphin (602) ⁴	0.063	0.005	0.051	0.006	NS
Sacramento R. across from Sherman Lake (704)	0.099	0.011	0.078	0.010	NS
Middle of Broad Slough, West end (804)	0.093	0.014	0.076	0.002	NS
Carquinez Strait, West of Benicia army dock (405) ⁵	0.031	0.004	0.037	0.011	NS
Suisun Bay off Chipps Island (508)	0.083	0.008	0.070	0.004	NS
Field Duplicate: Middle of Broad Slough, West end (804)	0.071	0.003	-	-	NA

	MSD	PMSD
One-way ANOVA	0.038	45.0
Two-way ANOVA	0.039	47.0

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$). Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable
3. This high conductivity sample was compared to the 12.53 mS/cm High EC control.
4. This high conductivity sample was compared to the 17.15 uS/cm High EC control.
5. This high conductivity sample was compared to the 24.14 mS/cm High EC control.

Table A88-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 8/22/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Montezuma Slough at Nurse Slough (609)	10450	24.9	7.02	9.0	12.1	0.010	0.0000
Suisun Bay, East of middle point (504)	12340	22.4	7.02	8.9	6.9	0.030	0.0001
Grizzly Bay at Dolphin (602)	17510	22.7	6.81	8.7	4.8	0.050	0.0001
Sacramento R. across from Sherman Lake (704)	1578	23.7	6.78	8.5	20.9	0.060	0.0002
Middle of Broad Slough, West end (804)	1960	22.6	6.64	8.6	8.1	0.030	0.0001
Carquinez Strait, West of Benicia army dock (405)	23850	22.6	6.73	8.9	10.1	0.140	0.0002
Suisun Bay off Chipps Island (508)	6780	22.3	6.32	8.9	9.5	0.020	0.0000
Field Duplicate: Middle of Broad Slough, West end (804)	-	-	-	-	8.1	-	-

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Table A88-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 08/23/2007 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 8/22/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	389	23.3	24.1	6.1	8.4	7.58	8.11	104	59	-
DIEPAMHR + organic matter	343	23.4	24.1	6.0	8.4	7.57	8.11	104	59	-
High EC Control @ 12.53 mS/cm	11710	23.5	24.1	6.2	8.3	7.46	8.01	-	-	-
High EC Control @ 17.15 mS/cm	15940	23.6	24.1	5.9	8.2	7.44	8.04	-	-	-
High EC Control @ 24.14 mS/cm	22745	23.6	24.0	6.0	8.1	7.49	8.09	2690	92	-
Montezuma Slough at Nurse Slough (609)	9955	23.6	24.0	6.2	8.9	7.68	7.92	1160	96	0.0003
Suisun Bay, East of middle point (504)	11525	23.7	24.0	6.4	8.5	7.56	7.84	1280	76	0.0007
Grizzly Bay at Dolphin (602)	15845	23.8	24.0	6.3	8.6	7.57	7.85	1840	84	0.0012
Sacramento R. across from Sherman Lake (704)	1570	23.7	24.0	5.9	8.4	7.67	8.07	196	64	0.0030
Middle of Broad Slough, West end (804)	1824	23.8	24.1	6.2	8.5	7.59	8.00	212	66	0.0013
Carquinez Strait, West of Benicia army dock (405)	22875	23.8	24.1	6.3	8.7	7.56	7.80	2680	94	0.0027
Suisun Bay off Chipps Island (508)	6595	23.8	24.1	6.1	8.7	7.52	7.91	732	70	0.0006
Field Duplicate: Middle of Broad Slough, West end (804)	1863	23.7	24.1	6.3	8.9	7.57	8.02	216	66	0.0013
DIEPAMHR + 25 ppb PBO	354	23.3	24.1	6.3	8.4	7.59	8.18	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	353	23.4	24.1	6.1	8.4	7.56	8.12	-	-	-
High EC Control @ 12.53 mS/cm + 25 ppb PBO	11555	23.3	24.1	6.3	8.7	7.48	8.01	-	-	-
High EC Control @ 17.15 mS/cm + 25 ppb PBO	15875	23.3	24.1	6.1	8.7	7.49	8.04	-	-	-
High EC Control @ 24.14 mS/cm + 25 ppb PBO	22425	23.3	24.1	5.9	8.5	7.53	8.04	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	9805	23.3	24.1	6.3	8.9	7.69	7.94	-	-	-
Suisun Bay, East of middle point (504) + 25 ppb PBO	11430	23.3	24.1	6.3	8.7	7.55	7.86	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	15800	23.3	24.1	6.2	8.5	7.58	7.86	-	-	-
Sacramento R. across from Sherman Lake (704) + 25 ppb PBO	1570	23.2	24.1	6.3	8.9	7.64	8.08	-	-	-
Middle of Broad Slough, West end (804) + 25 ppb PBO	1581	23.3	24.2	6.3	8.9	7.59	8.01	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	22490	23.4	24.2	6.3	8.9	7.61	7.80	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	6605	23.4	24.4	6.3	8.6	7.59	7.94	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A89-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 8/24/07 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 8/23/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	97	2.8	100	0.0	NS
DIEPAMHR + organic matter	100	0.0	100	0.0	NS
San Joaquin R. between Hog and Turner Cuts (910)	100	0.0	100	0.0	NS
San Joaquin R. West of Oulton Point (812)	98	2.5	100	0.0	NS
Old R., western arm at railroad bridge (902)	100	0.0	100	0.0	NS
Sacramento R. at tip of Grand Island (711) ³	88	2.5	98	2.5	NS
Sacramento R. Deep Water Channel, Light 55	100	0.0	100	0.0	NS
Old R. at mouth of Holland Cut (915)	100	0.0	95	2.9	NS

	MSD	PMSD
One-way ANOVA	7.4	7.4
Two-way ANOVA	7.6	7.6

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.054	0.010	0.043	0.002	NS
DIEPAMHR + organic matter	0.067	0.010	0.072	0.009	NS
San Joaquin R. between Hog and Turner Cuts (910)	0.081	0.013	0.082	0.013	NS
San Joaquin R. West of Oulton Point (812)	0.071	0.002	0.092	0.011	NS
Old R., western arm at railroad bridge (902)	0.119	0.020	0.109	0.011	NS
Sacramento R. at tip of Grand Island (711)	0.070	0.007	0.079	0.005	NS
Sacramento R. Deep Water Channel, Light 55	0.104	0.010	0.115	0.005	NS
Old R. at mouth of Holland Cut (915)	0.096	0.004	0.111	0.007	NS

	MSD	PMSD
One-way ANOVA	0.051	76.5
Two-way ANOVA	0.051	75.9

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$). Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable
3. This sample was found to show reduced survival because of the low MSD. This result was found both when the data were analyzed with Tukey's Multiple Comparison Procedure, and when the data were analyzed with USEPA standard statistics.

Table A89-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 8/23/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
San Joaquin R. between Hog and Turner Cuts (910)	416	25.4	7.12	6.4	6.62	0.040	0.0003
San Joaquin R. West of Oulton Point (812)	585	23.6	7.14	8.4	3.80	0.060	0.0004
Old R., western arm at railroad bridge (902)	735	25.5	8.07	9.2	4.53	0.000	0.0000
Sacramento R. at tip of Grand Island (711)	162	23.6	6.90	7.8	5.74	0.180	0.0007
Sacramento R. Deep Water Channel, Light 55	270	22.9	6.84	8.3	39.80	0.020	0.0001
Old R. at mouth of Holland Cut (915)	603	25.4	7.65	8.6	3.80	0.000	0.0000

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Table A89-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 08/24/2007 of samples collected by the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 8/23/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	349	23.6	24.2	6.1	8.2	7.54	8.00	104	59	-
DIEPAMHR + organic matter	343	23.2	24.2	6.3	8.3	7.52	8.15	104	59	-
San Joaquin R. between Hog and Turner Cuts (910)	407	23.2	24.2	6.1	8.7	7.59	7.71	100	77	0.001
San Joaquin R. West of Oulton Point (812)	568	23.3	24.2	6.1	8.8	7.48	7.86	96	63	0.002
Old R., western arm at railroad bridge (902)	705	23.2	24.2	6.2	8.8	7.55	8.38	108	61	0.000
Sacramento R. at tip of Grand Island (711)	166	23.3	24.2	5.8	8.6	7.49	7.72	58	66	0.004
Sacramento R. Deep Water Channel, Light 55	267	23.2	24.2	6.0	8.7	7.61	8.05	82	77	0.001
Old R. at mouth of Holland Cut (915)	586	23.3	24.2	6.1	8.7	7.44	8.09	96	62	0.000
DIEPAMHR + 25 ppb PBO	336	22.7	24.2	6.5	8.2	7.57	8.15	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	334	22.7	24.2	6.3	8.3	7.48	8.19	-	-	-
San Joaquin R. between Hog and Turner Cuts (910) + 25 ppb PBO	401	22.6	24.1	6.2	8.9	7.61	7.79	-	-	-
San Joaquin R. West of Oulton Point (812) + 25 ppb PBO	566	22.6	24.1	6.3	8.8	7.52	7.93	-	-	-
Old R., western arm at railroad bridge (902) + 25 ppb PBO	707	22.7	24.2	6.3	8.9	7.52	8.37	-	-	-
Sacramento R. at tip of Grand Island (711) + 25 ppb PBO	163	22.6	24.0	6.0	8.7	7.55	7.76	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	261	22.9	24.3	6.3	8.6	7.65	8.11	-	-	-
Old R. at mouth of Holland Cut (915) + 25 ppb PBO	620	22.9	24.3	6.0	8.6	7.48	8.02	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A90-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 9/5/07 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 9/4/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	95	2.9	95	2.9	NS
DIEPAMHR + organic matter	95	5.0	89	7.9	NS
High EC Control @ 12.97 mS/cm	93	4.8	79	7.1	NS
High EC Control @ 17.75 mS/cm	83	11.1	36	10.4	S (43%)
High EC Control @ 24.62 mS/cm	38	9.5	5	2.9	NS
Sacramento R. across from Sherman Lk. (704)	100	0.0	98	2.5	NS
Suisun Bay, East of middle point (504) ³	100	0.0	92	2.6	NS
Carquinez Strait, West of Benicia army dock (405) ⁵	13	4.8	15	8.7	NS
Suisun Bay, off Chipps Island (508)	100	0.0	95	2.9	NS
Grizzly Bay at dolphin (602) ⁴	92	2.7	77	6.1	NS
Montezuma Sl. At Nurse Sl. (609) ³	98	2.5	93	4.8	NS
Middle of Broad Slough, West end (804)	100	0.0	100	0.0	NS
Trip Blank (427)	98	2.5	-	-	NA

	MSD	PMSD
One-way ANOVA	24.5	25.8
Two-way ANOVA	28.8	30.3

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.022	0.005	0.046	0.004	NS
DIEPAMHR + organic matter	0.075	0.007	0.080	0.006	NS
High EC Control @ 12.97 mS/cm	0.070	0.007	0.066	0.006	NS
High EC Control @ 17.75 mS/cm	0.055	0.007	0.038	0.009	NS
High EC Control @ 24.62 mS/cm	0.082	0.015	0.045	0.005	NS
Sacramento R. across from Sherman Lk. (704)	0.076	0.007	0.065	0.005	NS
Suisun Bay, East of middle point (504) ³	0.069	0.008	0.055	0.008	NS
Carquinez Strait, West of Benicia army dock (405) ⁵	0.057	0.022	0.050	0.010	NS
Suisun Bay, off Chipps Island (508)	0.061	0.002	0.079	0.010	NS
Grizzly Bay at dolphin (602) ⁴	0.056	0.002	0.061	0.005	NS
Montezuma Sl. At Nurse Sl. (609) ³	0.073	0.004	0.073	0.005	NS
Middle of Broad Slough, West end (804)	0.073	0.002	0.086	0.007	NS
Trip Blank (427)	0.052	0.004	-	-	NA

	MSD	PMSD
One-way ANOVA	0.039	52.0
Two-way ANOVA	0.040	53.0

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$). Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.
3. This high conductivity sample was compared to the 12.97 mS/cm High EC control.
4. This high conductivity sample was compared to the 17.75 mS/cm High EC control.
5. This high conductivity sample was compared to the 24.62 mS/cm High EC control.

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Table A90-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 9/4/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Sacramento R. across from Sherman Lk. (704)	3180	22.5	7.51	8.1	12.6	0.040	0.000
Suisun Bay, East of middle point (504)	12610	22.2	7.66	8.3	12.3	0.060	0.001
Carquinez Strait, West of Benicia army dock (405)	23880	22.0	7.77	8.5	15.3	0.130	0.002
Suisun Bay, off Chipps Island (508)	7890	22.4	7.63	8.3	8.4	0.060	0.001
Grizzly Bay at dolphin (602)	17950	22.5	7.77	8.5	28.8	0.080	0.001
Montezuma Sl. At Nurse Sl. (609)	11930	23.6	7.59	7.6	18.3	0.090	0.001
Middle of Broad Slough, West end (804)	2444	23.4	7.53	8.0	8.9	0.100	0.001
Trip Blank (427)	336	23.6	8.21	7.9	0.3	0.010	0.001

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Table A90-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 09/05/2007 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 9/4/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	352	24.2	24.4	5.7	8.1	7.46	8.15	104	60	-
DIEPAMHR + organic matter	358	24.1	24.5	6.4	8.2	7.58	8.11	104	60	-
High EC Control @ 12.97 mS/cm	12050	24.2	24.5	6.7	7.8	7.55	8.10	1480	120	-
High EC Control @ 17.75 mS/cm	16600	24.3	24.5	6.6	8.7	7.58	8.09	2040	120	-
High EC Control @ 24.62 mS/cm	23205	24.2	24.5	6.4	8.1	7.66	8.11	2840	140	-
Sacramento R. across from Sherman Lk. (704)	3054	24.2	24.5	6.3	8.2	7.62	7.87	360	76	0.001
Suisun Bay, East of middle point (504)	12265	24.2	24.4	6.5	8.8	7.55	7.88	1400	100	0.002
Carquinez Strait, West of Benicia army dock (405)	23390	24.3	24.4	6.3	7.6	7.62	7.84	2800	140	0.003
Suisun Bay, off Chipps Island (508)	8065	24.0	24.4	6.6	8.4	7.66	7.93	960	90	0.002
Grizzly Bay at dolphin (602)	16960	24.3	24.4	6.1	7.9	7.54	7.88	1920	120	0.002
Montezuma Sl. At Nurse Sl. (609)	10920	23.8	24.5	6.3	8.2	7.69	7.82	1320	120	0.002
Middle of Broad Slough, West end (804)	2352	24.1	24.4	6.1	8.3	7.70	8.00	268	70	0.004
Trip Blank (427)	397	24.0	24.5	6.2	8.6	7.64	8.16	108	60	0.001
DIEPAMHR + 25 ppb PBO	357	24.2	24.4	6.1	8.2	7.61	8.14	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	362	22.8	24.4	6.3	8.1	7.62	8.19	-	-	-
High EC Control @ 12.97 mS/cm + 25 ppb PBO	12000	23.1	24.4	6.6	8.3	7.59	8.04	-	-	-
High EC Control @ 17.75 mS/cm + 25 ppb PBO	16410	22.9	24.6	6.5	8.1	7.60	8.06	-	-	-
High EC Control @ 24.62 mS/cm + 25 ppb PBO	22685	22.7	24.5	6.6	8.9	7.69	8.10	-	-	-
Sacramento R. across from Sherman Lk. (704) + 25 ppb PBO	3079.5	22.6	24.5	6.7	8.5	7.74	8.05	-	-	-
Suisun Bay, East of middle point (504) + 25 ppb PBO	12010	22.7	24.5	6.5	8.2	7.59	7.93	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	22750	22.6	24.5	6.1	8.3	7.59	7.85	-	-	-
Suisun Bay, off Chipps Island (508) + 25 ppb PBO	7750	22.5	24.5	6.5	8.8	7.65	7.96	-	-	-
Grizzly Bay at dolphin (602) + 25 ppb PBO	16350	22.5	24.5	6.1	8.6	7.56	7.87	-	-	-
Montezuma Sl. At Nurse Sl. (609) + 25 ppb PBO	10910	22.5	24.5	6.5	8.8	7.75	7.76	-	-	-
Middle of Broad Slough, West end (804) + 25 ppb PBO	2380	22.5	24.6	6.4	8.9	7.13	7.75	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A91-1. Survival of *H. azteca* in a Phase I Toxicity Identification Evaluation initiated on 9/12/07 examining an ambient water sample collected at POD site 405 by the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 9/04/07.

Treatment	Survival (%)									
	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Day 8	Day 9	Day 10
DIEPAMHR	100	100	100	100	100	100	100	100	100	100
DIEPAMHR (ECA) @ 24.62 mS/cm	100	100	93	93	90	83	83	83	80	80
DIEPAMHR + MeOH @ 0.5%	100	100	100	100	87	87	87	87	87	87
DIEPAMHR + eluate addback @ 3x ²	95	95	95	90	60	50	50	50	50	43
DIEPAMHR (ECA) + 120 mg/L EDTA	100	100	97	97	30	13	7	7	7	7
DIEPAMHR (ECA) + 30 mg/L EDTA	100	97	97	97	53	53	43	40	37	37
DIEPAMHR (ECA) + 7.5 mg/L EDTA	100	100	100	97	83	69	66	62	62	62
DIEPAMHR (ECA) + 2 mg/L STS	100	100	97	97	83	73	59	59	59	59
DIEPAMHR (ECA) + 0.5 mg/L STS	100	100	100	100	52	42	39	28	28	25
DIEPAMHR (ECA) + 0.125 mg/L STS	100	100	97	93	74	56	48	48	48	45
DIEPAMHR (ECA) air stripped	100	100	100	100	60	60	56	56	53	53
DIEPAMHR C8 Blank	100	100	100	100	97	97	97	97	97	97
POD 405 090407	100	100	97	93	93	93	93	90	90	87
POD 405 + 120 mg/L EDTA	100	100	76	76	76	76	76	70	70	70
POD 405 + 30 mg/L EDTA	100	100	100	100	100	100	95	86	86	86
POD 405 + 7.5 mg/L EDTA	100	100	93	93	90	90	90	90	90	90
POD 405 + 2 mg/L STS	100	96	93	93	86	82	79	75	71	71
POD 405 + 0.5 mg/L STS	100	100	97	93	93	93	93	93	90	86
POD 405 + 0.125 mg/L STS	100	100	97	97	88	88	88	88	85	81
POD 405 air stripped	100	100	93	93	93	86	83	78	78	78
POD 405 C8 Rinsate	100	93	89	89	82	82	79	71	71	68

1. Highlighted cells indicate 50% or greater mortality.
2. Sample was eluted through a C8 column on 9/10/07.

Table A92-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 9/6/07 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 9/5/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	94	3.4	95	5.0	NS
DIEPAMHR + organic matter	81	9.6	82	8.0	NS
Old R., western arm at railroad bridge (902)	95	2.9	97	3.1	NS
San Joaquin R. between Hog and Turner Cuts (910)	100	0.0	98	2.5	NS
Sacramento R. Deep Water Channel, Light 55	100	0.0	93	7.5	NS
Sacramento R. at tip of Grand Island (711)	83	6.7	87	4.7	NS
Old R. at mouth of Holland Cut (915)	98	2.5	90	10.0	NS
San Joaquin R., West of Oulton Pt. (812)	98	2.5	98	2.5	NS
Sacramento R. at Hood DWR Station	78	7.5	98	2.5	NS
Trip Blank (527)	100	0.0	-	-	NA
Field Dup.: Sacramento R. at Hood DWR Station	88	4.8	-	-	NA
Bottle Blank (227)	92	4.8	-	-	NA

	MSD	PMSD
One-way ANOVA	23.8	29.4
Two-way ANOVA	28.0	34.6

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.028	0.002	0.050	0.007	NS
DIEPAMHR + organic matter	0.040	0.007	0.061	0.002	NS
Old R., western arm at railroad bridge (902)	0.057	0.005	0.066	0.005	NS
San Joaquin R. between Hog and Turner Cuts (910)	0.066	0.011	0.047	0.006	NS
Sacramento R. Deep Water Channel, Light 55	0.074	0.005	0.090	0.004	NS
Sacramento R. at tip of Grand Island (711)	0.044	0.009	0.056	0.003	NS
Old R. at mouth of Holland Cut (915)	0.050	0.002	0.052	0.009	NS
San Joaquin R., West of Oulton Pt. (812)	0.067	0.007	0.053	0.008	NS
Sacramento R. at Hood DWR Station	0.045	0.003	0.063	0.006	NS
Trip Blank (527)	0.051	0.005	-	-	NA
Field Dup.: Sacramento R. at Hood DWR Station	0.057	0.005	-	-	NA
Bottle Blank (227)	0.043	0.003	-	-	NA

	MSD	PMSD
One-way ANOVA	0.029	72.5
Two-way ANOVA	0.032	79.2

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$). Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable

Table A92-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 9/5/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Old R., western arm at railroad bridge (902)	830.0	23.5	8.24	8.3	6.1	0.020	0.001
San Joaquin R. between Hog and Turner Cuts (910)	475.8	24.2	7.31	6.1	8.7	0.050	0.000
Sacramento R. Deep Water Channel, Light 55	265.3	22.9	7.60	7.9	40.0	0.030	0.001
Sacramento R. at tip of Grand Island (711)	173.7	22.4	7.10	7.8	17.9	0.130	0.001
Old R. at mouth of Holland Cut (915)	623.0	23.8	7.78	7.8	4.8	0.000	0.000
San Joaquin R., West of Oulton Pt. (812)	393.9	22.7	7.55	7.8	5.6	0.060	0.001
Sacramento R. at Hood DWR Station	169.6	23.7	7.48	7.9	13.8	0.100	0.001
Trip Blank (527)	371.4	23.4	8.25	8.1	0.7	0.000	0.000
Field Dup.: Sacramento R. at Hood DWR Station	166.2	23.7	7.48	7.6	12.5	0.090	0.001
Bottle Blank (227)	-	-	-	-	0.4	0.000	-

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Table A92-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 09/06/2007 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 9/5/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	359.2	23.2	23.8	5.8	8.1	7.43	8.04	104	56	-
DIEPAMHR + Organic Matter	354.9	22.9	24.0	5.4	8.3	7.57	8.05	104	56	-
Old R., western arm at railroad bridge (902)	805.0	23.2	23.7	6.4	8.4	7.52	8.10	124	68	0.001
San Joaquin R. between Hog and Turner Cuts (910)	486.5	23.0	23.8	6.1	8.2	7.59	7.74	112	84	0.001
Sacramento R. Deep Water Channel, Light 55	289.7	23.0	23.9	5.9	8.2	7.59	8.04	88	82	0.001
Sacramento R. at tip of Grand Island (711)	173.9	22.9	23.8	6.2	8.3	7.52	7.89	52	68	0.004
Old R. at mouth of Holland Cut (915)	689.5	22.9	23.7	6.3	8.4	7.53	7.92	116	68	0.000
San Joaquin R., West of Oulton Pt. (812)	404.9	23.0	23.7	6.0	8.5	7.51	7.83	92	68	0.002
Sacramento R. at Hood DWR Station	166.0	22.9	23.8	6.2	8.3	7.46	7.84	64	70	0.003
Trip Blank (527)	335.4	22.6	23.7	5.9	8.5	7.39	8.09	100	58	0.000
Field Dup.: Sacramento R. at Hood DWR Station	167.7	22.8	23.8	5.5	8.7	7.37	7.94	60	68	0.003
Bottle Blank (227)	338.9	22.9	23.7	6.0	8.4	7.41	8.12	108	60	0.000
DIEPAMHR + 25 ppb PBO	338.6	22.1	23.9	6.3	8.2	7.44	8.08	-	-	-
DIEPAMHR + Organic Matter 25 ppb PBO	336.2	22.0	23.9	6.3	8.2	7.47	8.07	-	-	-
Old R., western arm at railroad bridge (902) + 25 ppb PBO	770.0	22.0	23.9	6.5	8.6	7.54	8.15	-	-	-
San Joaquin R. between Hog and Turner Cuts (910) + 25 ppb PBO	451.7	22.1	23.8	6.6	8.4	7.56	7.93	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	266.8	22.0	23.9	6.3	8.7	7.64	8.20	-	-	-
Sacramento R. at tip of Grand Island (711) + 25 ppb PBO	162.7	22.2	23.8	6.0	8.3	7.44	7.97	-	-	-
Old R. at mouth of Holland Cut (915) + 25 ppb PBO	684.0	22.1	24.1	6.6	8.7	7.62	8.02	-	-	-
San Joaquin R., West of Oulton Pt. (812) + 25 ppb PBO	395.9	22.0	23.9	6.3	8.6	7.60	7.97	-	-	-
Sacramento R. at Hood DWR Station + 25 ppb PBO	166.8	22.0	24.0	6.1	8.8	7.45	7.93	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A93-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 9/20/07 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 9/19/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	95	2.9	100	0.0	NS
DIEPAMHR + organic matter	88	7.5	83	2.5	NS
High EC Control @ 13.15 mS/cm + organic matter	93	2.5	83	7.5	NS
High EC Control @ 16.01 mS/cm + organic matter	84	5.2	77	7.8	NS
High EC Control @ 24.23 mS/cm + organic matter	54	9.1	38	9.3	NS
Suisun Bay, East of middle point (504) ³	90	7.1	70	20.5	NS
Carquinez Strait, West of Benicia army dock (405) ⁵	37	5.8	13	4.8	NS
Sacramento R. across from Sherman Lk. (704)	98	2.5	95	2.9	NS
Middle of Broad Sl., West end (804)	98	2.5	97	2.8	NS
Suisun Bay off Chipps Island (508)	98	2.5	90	5.8	NS
Montezuma Sl. at Nurse Sl. (609) ³	92	4.8	93	4.8	NS
Grizzly Bay at dolphin (602) ⁴	87	7.1	90	4.1	NS
Trip Blank (527)	92	2.6	-	-	NA

	MSD	PMSD
One-way ANOVA	26.4	30.2
Two-way ANOVA	36.5	41.7

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.047	0.002	0.035	0.002	NS
DIEPAMHR + organic matter	0.055	0.007	0.038	0.002	NS
High EC Control @ 13.15 mS/cm + organic matter	0.058	0.006	0.035	0.006	NS
High EC Control @ 16.01 mS/cm + organic matter	0.058	0.004	0.035	0.010	NS
High EC Control @ 24.23 mS/cm + organic matter	0.033	0.002	0.033	0.003	NS
Suisun Bay, East of middle point (504) ³	0.055	0.005	0.036	0.008	NS
Carquinez Strait, West of Benicia army dock (405) ⁵	0.026	0.008	0.042	0.009	NS
Sacramento R. across from Sherman Lk. (704)	0.050	0.004	0.060	0.006	NS
Middle of Broad Sl., West end (804)	0.063	0.007	0.047	0.004	NS
Suisun Bay off Chipps Island (508)	0.051	0.011	0.046	0.003	NS
Montezuma Sl. at Nurse Sl. (609) ³	0.054	0.003	0.030	0.004	NS
Grizzly Bay at dolphin (602) ⁴	0.054	0.004	0.024	0.002	S (44%)
Trip Blank (527)	0.046	0.004	-	-	NA

	MSD	PMSD
One-way ANOVA	0.028	51.1
Two-way ANOVA	0.030	54.4

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. This high conductivity sample was compared to the 13.15 mS/cm High EC control.

4. This high conductivity sample was compared to the 16.01 mS/cm High EC control.

5. This high conductivity sample was compared to the 24.23 mS/cm High EC control.

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Table A93-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 9/19/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Suisun Bay, East of middle point (504)	10190	18.5	7.10	9.1	9.5	0.050	0.000
Carquinez Strait, West of Benicia army dock (405)	21750	18.1	7.20	9.1	19.6	0.050	0.000
Sacramento R. across from Sherman Lk. (704)	1708	19.1	7.00	8.9	15.2	0.020	0.000
Middle of Broad Sl., West end (804)	1511	19.8	7.48	8.9	6.3	0.000	0.000
Suisun Bay off Chipps Island (508)	5700	18.4	6.80	8.9	10.0	0.030	0.000
Montezuma Sl. at Nurse Sl. (609)	12340	19.0	7.30	8.8	27.3	0.000	0.000
Grizzly Bay at dolphin (602)	15440	17.9	7.10	9.1	21.9	0.040	0.000
Trip Blank (527)	343	21.4	8.06	8.5	0.2	0.000	0.000

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Table A93-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 09/20/2007 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 9/19/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	338	22.0	22.2	6.5	8.4	7.52	8.13	104	58	-
DIEPAMHR + organic matter	327	22.2	22.5	6.0	8.6	7.59	8.11	104	58	-
High EC Control @ 13.15 mS/cm + organic matter	11950	22.2	22.7	6.3	8.6	7.46	7.93	1400	74	-
High EC Control @ 16.01 mS/cm + organic matter	14360	22.4	22.6	6.6	8.8	7.55	7.94	1760	74	-
High EC Control @ 24.23 mS/cm + organic matter	22205	22.4	22.7	6.8	8.6	7.64	7.93	2760	88	-
Suisun Bay, East of middle point (504)	9650	22.5	22.8	6.5	8.5	7.51	7.91	1220	78	0.001
Carquinez Strait, West of Benicia army dock (405)	21945	22.4	22.5	6.1	8.4	7.58	7.82	2720	94	0.001
Sacramento R. across from Sherman Lk. (704)	1738	22.5	22.5	6.8	8.7	7.61	8.08	220	72	0.001
Middle of Broad Sl., West end (804)	8012	22.4	22.6	6.5	8.4	7.54	8.12	188	80	0.000
Suisun Bay off Chipps Island (508)	5530	22.1	22.6	6.4	8.7	7.51	7.90	840	76	0.001
Montezuma Sl. at Nurse Sl. (609)	11610	22.3	22.4	6.7	8.6	7.65	7.81	1600	96	0.000
Grizzly Bay at dolphin (602)	14670	22.5	22.6	6.8	8.5	7.62	7.85	2080	82	0.001
Trip Blank (527)	433	22.7	22.7	6.7	8.6	7.51	8.19	104	60	0.000
DIEPAMHR + 25 ppb PBO	413	22.3	22.4	6.6	8.4	7.58	8.13	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	354	22.4	22.6	6.3	8.4	7.42	8.12	-	-	-
High EC Control @ 13.15 mS/cm + organic matter + 25 ppb PBO	11785	22.6	22.8	6.6	8.6	7.46	7.93	-	-	-
High EC Control @ 16.01 mS/cm + organic matter + 25 ppb PBO	14760	22.5	22.8	6.8	8.9	7.53	7.94	-	-	-
High EC Control @ 24.23 mS/cm + organic matter + 25 ppb PBO	21815	22.5	22.6	6.9	8.5	7.66	7.93	-	-	-
Suisun Bay, East of middle point (504) + 25 ppb PBO	9750	22.6	22.7	6.8	8.4	7.54	7.95	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	21975	22.5	22.7	6.7	8.6	7.58	7.79	-	-	-
Sacramento R. across from Sherman Lk. (704) + 25 ppb PBO	1770.5	22.7	22.7	6.7	8.5	7.56	8.13	-	-	-
Middle of Broad Sl., West end (804) + 25 ppb PBO	1524.5	22.6	22.6	6.6	8.4	7.57	8.11	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	5595	22.1	22.7	6.7	8.6	7.57	7.91	-	-	-
Montezuma Sl. at Nurse Sl. (609) + 25 ppb PBO	11605	22.5	22.6	6.8	8.5	7.61	7.91	-	-	-
Grizzly Bay at dolphin (602) + 25 ppb PBO	14370	22.5	22.5	6.5	8.5	7.54	7.85	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A94-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 9/20/07 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 9/21/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	92	2.6	92	4.8	NS
DIEPAMHR + organic matter	98	2.5	91	6.4	NS
Sacramento R. at tip of Grand Island (711)	93	4.8	88	5.6	NS
Sacramento R. Deep Water Channel, Light 55	92	4.9	95	5.0	NS
Old R. at mouth of Holland Cut (915)	100	0.0	98	2.5	NS
San Joaquin R. between Hog and Turner Cuts (910)	97	3.1	95	2.9	NS
Old R., western arm at railroad bridge (902)	94	5.6	93	4.8	NS
San Joaquin R., West of Oulton Point (812)	98	2.5	100	0.0	NS
Sacramento R. at Hood DWR Station	74	11.7	78	4.3	NS
Trip Blank (427)	98	2.5	-	-	NA
Bottle Blank (227)	92	4.8	-	-	NA

	MSD	PMSD
One-way ANOVA	24.5	25.1
Two-way ANOVA	24.7	25.3

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.034	0.004	0.036	0.002	NS
DIEPAMHR + organic matter	0.046	0.006	0.046	0.006	NS
Sacramento R. at tip of Grand Island (711)	0.049	0.002	0.048	0.003	NS
Sacramento R. Deep Water Channel, Light 55	0.060	0.007	0.057	0.002	NS
Old R. at mouth of Holland Cut (915)	0.080	0.005	0.052	0.007	NS
San Joaquin R. between Hog and Turner Cuts (910)	0.063	0.006	0.060	0.005	NS
Old R., western arm at railroad bridge (902)	0.072	0.003	0.055	0.006	NS
San Joaquin R., West of Oulton Point (812)	0.067	0.006	0.056	0.006	NS
Sacramento R. at Hood DWR Station	0.054	0.003	0.035	0.004	NS
Trip Blank (427)	0.045	0.003	-	-	NA
Bottle Blank (227)	0.029	0.003	-	-	NA

	MSD	PMSD
One-way ANOVA	0.023	50.7
Two-way ANOVA	0.026	56.0

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.
Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

Table A94-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 9/20/07 - 9/21/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Sacramento R. at tip of Grand Island (711)	170.9	18.6	7.40	8.5	6.1	0.180	0.002
Sacramento R. Deep Water Channel, Light 55	286.4	19.0	7.50	8.6	42.2	0.020	0.000
Old R. at mouth of Holland Cut (915)	684.0	19.4	7.40	8.7	4.5	0.000	0.000
San Joaquin R. between Hog and Turner Cuts (910)	454.5	20.5	7.30	7.3	10.5	0.000	0.000
Old R., western arm at railroad bridge (902)	620.0	19.2	8.10	8.9	4.3	0.000	0.000
San Joaquin R., West of Oulton Point (812)	439.7	19.1	7.50	8.7	7.6	0.040	0.000
Sacramento R. at Hood DWR Station	157.1	19.5	8.76	8.6	14.1	0.170	0.029
Trip Blank (427)	310.4	19.7	8.15	8.7	2.0	0.000	0.000
Bottle Blank (227)	-	-	-	-	0.4	-	-

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Table A94-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 08/24/2007 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 8/23/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	342.6	22.2	22.4	6.0	8.5	7.48	8.07	104	58	-
DIEPAMHR + organic matter	341.2	22.1	22.7	5.9	8.5	7.35	8.14	104	58	-
Sacramento R. at tip of Grand Island (711)	180.9	22.2	23.0	5.4	8.6	7.44	7.84	60	70	0.006
Sacramento R. Deep Water Channel, Light 55	265.1	22.1	22.2	5.8	8.7	7.58	8.04	88	82	0.001
Old R. at mouth of Holland Cut (915)	651.0	22.1	22.8	6.2	8.6	7.55	8.11	108	68	0.000
San Joaquin R. between Hog and Turner Cuts (910)	443.9	22.0	22.9	6.0	8.5	7.59	7.91	112	84	0.000
Old R., western arm at railroad bridge (902)	672.0	22.1	23.1	6.1	8.5	7.51	8.25	108	68	0.000
San Joaquin R., West of Oulton Point (812)	482.9	21.9	23.0	5.9	8.7	7.54	8.16	92	70	0.002
Sacramento R. at Hood DWR Station	163.2	21.9	22.7	5.2	8.7	7.34	7.62	68	66	0.003
Trip Blank (427)	340.8	21.9	22.5	6.6	8.5	7.56	8.15	104	58	0.000
Bottle Blank (227)	337.5	21.9	23.1	6.5	8.4	7.52	8.05	116	60	-
DIEPAMHR + 25 ppb PBO	339.9	21.9	23.2	6.3	8.4	7.53	8.14	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	337.0	21.9	23.2	5.8	8.6	7.48	8.16	-	-	-
Sacramento R. at tip of Grand Island (711) + 25 ppb PBO	176.6	21.9	23.2	5.8	8.7	7.49	7.92	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	269.3	21.9	23.3	5.9	8.5	7.65	8.12	-	-	-
Old R. at mouth of Holland Cut (915) + 25 ppb PBO	666.0	21.9	23.4	6.1	8.6	7.56	8.16	-	-	-
San Joaquin R. between Hog and Turner Cuts (910) + 25 ppb PBO	437.9	21.9	23.5	6.1	8.4	7.61	7.98	-	-	-
Old R., western arm at railroad bridge (902) + 25 ppb PBO	671.5	22.0	23.3	5.9	8.6	7.53	8.24	-	-	-
San Joaquin R., West of Oulton Point (812) + 25 ppb PBO	485.6	21.9	23.4	6.2	8.5	7.56	8.06	-	-	-
Sacramento R. at Hood DWR Station + 25 ppb PBO	161.0	21.9	23.3	5.1	8.6	7.35	7.55	116	60	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A95-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 10/4/07 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 10/2/07 - 10/3/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	97	2.8	98	2.5	NS
DIEPAMHR + organic matter	100	0.0	100	0.0	NS
Sacramento R. Deep Water Channel, Light 55	100	0.0	98	2.5	NS
Sacramento R. at tip of Grand Island (711)	100	0.0	100	0.0	NS
San Joaquin R., West of Oulton Point (812)	100	0.0	98	2.5	NS
San Joaquin R. between Hog and Turner Cut (910)	100	0.0	100	0.0	NS
Old R. at mouth of Holland Cut (915)	100	0.0	100	0.0	NS
Old R., western arm at railroad bridge (902)	100	0.0	100	0.0	NS
Sacramento R. at Hood DWR Station	43	7.3	89	4.5	S (207%)
Field Dup: Sacramento R. at tip of Grand Island (711)	100	0.0	-	-	NA
Bottle Blank	100	0.0	-	-	NA

	MSD	PMSD
One-way ANOVA	11.6	11.6
Two-way ANOVA	12.3	12.3

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.046	0.006	0.038	0.002	NS
DIEPAMHR + organic matter	0.060	0.008	0.068	0.010	NS
Sacramento R. Deep Water Channel, Light 55	0.064	0.006	0.074	0.005	NS
Sacramento R. at tip of Grand Island (711)	0.067	0.009	0.075	0.001	NS
San Joaquin R., West of Oulton Point (812)	0.075	0.009	0.082	0.006	NS
San Joaquin R. between Hog and Turner Cut (910)	0.055	0.008	0.083	0.005	NS
Old R. at mouth of Holland Cut (915)	0.069	0.004	0.072	0.008	NS
Old R., western arm at railroad bridge (902)	0.068	0.006	0.060	0.006	NS
Sacramento R. at Hood DWR Station	0.093	0.019	0.062	0.006	NS
Field Dup: Sacramento R. at tip of Grand Island (711)	0.076	0.009	-	-	NA
Bottle Blank	0.046	0.003	-	-	NA

	MSD	PMSD
One-way ANOVA	0.044	72.5
Two-way ANOVA	0.041	67.9

- Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$). Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
- NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

Table A95-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 10/2/07 - 10/3/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Sacramento R. Deep Water Channel, Light 55	177	18.2	7.70	8.9	14.8	0.17	0.003
Sacramento R. at tip of Grand Island (711)	158	17.8	7.67	8.9	7.3	0.25	0.004
San Joaquin R., West of Oulton Point (812)	500	18.1	7.87	9.1	6.0	0.07	0.002
San Joaquin R. between Hog and Turner Cut (910)	528	19.7	7.65	7.2	7.2	0.06	0.001
Old R. at mouth of Holland Cut (915)	595	18.6	7.97	9.2	3.8	0.02	0.001
Old R., western arm at railroad bridge (902)	651	18.5	7.88	9.4	7.6	0.03	0.001
Sacramento R. at Hood DWR Station	142	18.6	7.23	8.7	6.9	0.31	0.002
Field Dup: Sacramento R. at tip of Grand Island (711)	158	17.8	7.67	8.9	7.6	0.23	0.003
Bottle Blank	-	-	-	-	0.3	-	-

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Table A95-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 10/04/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 10/02/07- 10/03/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	350	22.2	22.2	6.5	8.6	7.50	8.19	102	58	-
DIEPAMHR + organic matter	337	22.1	22.5	6.7	8.6	7.50	8.06	102	58	-
Sacramento R. Deep Water Channel, Light 55	183	22.2	23.0	6.0	8.5	7.49	7.82	66	70	0.005
Sacramento R. at tip of Grand Island (711)	164	22.1	23.1	6.3	8.5	7.50	7.91	54	63	0.009
San Joaquin R., West of Oulton Point (812)	488	22.2	23.1	6.4	8.6	7.53	7.87	90	68	0.002
San Joaquin R. between Hog and Turner Cut (910)	508	22.5	23.0	6.5	8.6	7.65	7.79	122	93	0.002
Old R. at mouth of Holland Cut (915)	575	22.4	23.2	6.8	8.8	7.58	8.01	100	71	0.001
Old R., western arm at railroad bridge (902)	615	22.3	22.9	6.6	8.8	7.58	8.08	100	70	0.002
Sacramento R. at Hood DWR Station	144	22.4	22.9	6.2	8.7	7.29	7.73	60	61	0.008
Field Dup: Sacramento R. at tip of Grand Island (711)	166	22.4	22.9	6.4	8.8	7.51	7.86	56	64	0.007
Bottle Blank	337	22.3	22.9	6.8	8.7	7.52	8.04	100	58	0.000
DIEPAMHR + 25 ppb PBO	333	22.5	22.7	7.0	8.4	7.54	8.07	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	328	22.4	22.8	6.8	8.6	7.46	8.04	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	182	22.3	22.8	6.2	8.7	7.50	7.89	-	-	-
Sacramento R. at tip of Grand Island (711) + 25 ppb PBO	163	22.4	22.8	6.3	8.7	7.45	7.87	-	-	-
San Joaquin R., West of Oulton Point (812) + 25 ppb PBO	490	22.4	22.4	6.7	8.9	7.52	7.91	-	-	-
San Joaquin R. between Hog and Turner Cut (910) + 25 ppb PBO	501	22.3	22.7	6.9	8.9	7.72	7.86	-	-	-
Old R. at mouth of Holland Cut (915) + 25 ppb PBO	572	22.3	22.5	6.5	8.8	7.55	8.02	-	-	-
Old R., western arm at railroad bridge (902) + 25 ppb PBO	602	22.4	22.7	6.7	8.8	7.57	8.06	-	-	-
Sacramento R. at Hood DWR Station + 25 ppb PBO	144	22.4	23.1	6.3	8.6	7.29	7.73	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A96-1. Survival of *H. azteca* in a Phase I Toxicity Identification Evaluation initiated on 10/21/07 examining causes of toxicity in an ambient water column sample collected at the HOOD DWR Station on the Sacramento River by the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 10/02/07.¹

Treatment	Mean Survival (%) ²									
	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Day 8	Day 9	Day 10
DIEPAMHR	100	100	100	100	100	100	83	83	83	83
DIEPAMHR (HA) @ 60 mg/L	100	100	100	100	100	100	100	100	100	100
DIEPAMHR (HA) + MeOH @ 0.5%	100	100	100	100	100	97	90	90	90	90
DIEPAMHR (HA) + eluate addback @ 3x	100	100	100	100	100	100	100	90	90	90
DIEPAMHR (HA) + eluate addback @ 1x	100	100	100	100	100	100	100	97	93	87
DIEPAMHR C8 Blank	100	100	100	100	100	100	100	100	100	100
POD Hood 100207	97	97	97	97	97	97	97	97	97	97
POD Hood C8 Rinsate	100	100	100	100	100	100	100	100	100	100

1. Sample was treated through Varian C8 column on 10/19/07.
2. Highlighted cells indicate less than 50% survival of test organisms.

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Table A96-2. Water chemistry measured during a 10 day *H. azteca* Phase I Toxicity Identification Evaluation initiated on 10/21/07 examining causes of toxicity in an ambient water column sample collected 10/02/07 at the Hood DWR Station on the Sacramento River.

Treatment	Mean EC (uS/cm)	Min Temp	Max Temp	Min DO	Max DO	Min pH	Max pH
DIEPAMHR	370.9	21.3	21.3	6.9	8.6	7.55	7.65
DIEPAMHR (HA) @ 60 mg/L	235.9	21.0	21.0	6.5	8.6	7.27	7.5
DIEPAMHR (HA) + MeOH @ 0.5%	212.5	21.5	21.5	1.2	8.6	6.90	7.09
DIEPAMHR (HA) + eluate addback @ 3x	218.6	21.3	21.3	1.2	8.6	6.92	7.12
DIEPAMHR (HA) + eluate addback @ 1x	223.5	21.3	21.3	5.4	8.6	7.06	7.37
DIEPAMHR C8 Blank	379.4	21.4	21.4	6.8	8.6	7.55	7.63
POD Hood 100207	178.9	21.6	21.6	6.4	10.3	7.48	7.58
POD Hood C8 Rinsate	182.9	21.8	21.8	7.0	10.3	7.62	7.64

Table A97-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 10/5/07 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 10/4/07 - 10/5/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	100	0.0	100	0.0	NS
DIEPAMHR + organic matter	100	0.0	100	0.0	NS
High EC Control @ 13.99 mS/cm + organic matter	100	0.0	98	2.5	NS
High EC Control @ 18.51 mS/cm + organic matter	100	0.0	95	3.1	NS
High EC Control @ 25.40 mS/cm + organic matter	98	2.5	97	2.8	NS
Carquinez Strait, West of Benicia army dock (405) ⁵	76	5.0	77	4.6	NS
Grizzly Bay at Dolphin (602) ⁴	100	0.0	95	5.0	NS
Sacramento R. across from Sherman Lake (704)	98	2.5	98	2.5	NS
Suisun Bay off Chipps Island (508)	90	4.1	100	0.0	NS
Montezuma Sl. At Nurse Sl. (609) ³	100	0.0	100	0.0	NS
Middle of Broad Sl., West end (804)	98	2.5	100	0.0	NS
Pachecco Creek Outflow (409) ^{5,6}	78	9.5	25	6.2	S (32%)
Suisun Bay, East of middle point (504) ³	100	0.0	98	2.5	NS
Trip Blank (427)	100	0.0	-	-	NA

	MSD	PMSD
One-way ANOVA	16.5	16.5
Two-way ANOVA	17.6	17.6

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.042	0.009	0.042	0.004	NS
DIEPAMHR + organic matter	0.070	0.006	0.078	0.009	NS
High EC Control @ 13.99 mS/cm + organic matter	0.044	0.009	0.058	0.007	NS
High EC Control @ 18.51 mS/cm + organic matter	0.034	0.005	0.047	0.009	NS
High EC Control @ 25.40 mS/cm + organic matter	0.024	0.007	0.038	0.005	NS
Carquinez Strait, West of Benicia army dock (405) ⁵	0.050	0.006	0.048	0.003	NS
Grizzly Bay at Dolphin (602) ⁴	0.038	0.006	0.042	0.006	NS
Sacramento R. across from Sherman Lake (704)	0.040	0.011	0.050	0.007	NS
Suisun Bay off Chipps Island (508)	0.050	0.004	0.049	0.006	NS
Montezuma Sl. At Nurse Sl. (609) ³	0.050	0.004	0.035	0.010	NS
Middle of Broad Sl., West end (804)	0.077	0.005	0.077	0.007	NS
Pachecco Creek Outflow (409) ^{5,6}	0.039	0.004	0.110	0.033	NS
Suisun Bay, East of middle point (504) ³	0.052	0.005	0.070	0.010	NS
Trip Blank (427)	0.040	0.007	-	-	NA

	MSD	PMSD
One-way ANOVA	0.033	47.5
Two-way ANOVA	0.051	73.8

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.
3. This high conductivity sample was compared to the 13.99 mS/cm High EC control.
4. This high conductivity sample was compared to the 18.51 mS/cm High EC control.
5. This high conductivity sample was compared to the 25.40 mS/cm High EC control.
6. This was a permethrin spiked sample. See Table A97-2.

Table A97-2. Summary of a blind spike in a 10-day *H. azteca* water column toxicity test initiated on 10/5/07 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 10/4/07 - 10/5/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	100	0.0	100	0.0	NS
DIEPAMHR + organic matter	100	0.0	100	0.0	NS
High EC Control @ 18.51 mS/cm + organic matter	100	0.0	95	3.1	NS
High EC Control @ 25.40 mS/cm + organic matter	98	2.5	97	2.8	NS
Permethrin Spike @ 23 mS/cm ³	78	9.5	25	6.2	S (32%)

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.042	0.009	0.042	0.004	NS
DIEPAMHR + organic matter	0.070	0.006	0.078	0.009	NS
High EC Control @ 18.51 mS/cm + organic matter	0.034	0.005	0.047	0.009	NS
High EC Control @ 25.40 mS/cm + organic matter	0.024	0.007	0.038	0.005	NS
Permethrin Spike @ 23 mS/cm ^{3,4}	0.039	0.004	0.110	0.033	S (282%)

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$). Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.
3. This high conductivity sample was compared to the 25.40 mS/cm High EC control.
4. The significant increase in weight with PBO addition may be an artifact of scale imprecision, since 3 of the replicates of this treatment showed low weights, and 1 replicate containing 1 animal was found to be very heavy.

Table A97-3. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 10/4/07 - 10/5/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Carquinez Strait, West of Benicia army dock (405)	23670	17.3	7.71	9.5	31.0	0.1	0.0011
Grizzly Bay at Dolphin (602)	17920	17.3	7.71	9.2	28.6	0.11	0.0012
Sacramento R. across from Sherman Lake (704)	2733	18.1	7.84	9.5	19.3	0.06	0.0012
Suisun Bay off Chipps Island (508)	6410	17.6	7.77	9.2	6.7	0.07	0.0010
Montezuma Sl. At Nurse Sl. (609)	13390	18.2	7.58	8.8	10.3	0.01	0.0001
Middle of Broad Sl., West end (804)	1972	18.4	7.92	9.4	8.2	0.05	0.0012
Suisun Bay, East of middle point (504)	10900	17.4	7.75	9.1	7.7	0.06	0.0008
Trip Blank (427)	337	20.7	8.12	8.7	0.1	0.01	0.0005

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Table A97-4. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 10/5/07 of samples collected by the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 10/4/07 - 10/5/07.

Treatment	Laboratory Chemistry						Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹	
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH				Max pH
DIEPAMHR	357	21.6	22.6	6.8	8.3	7.59	8.23	102	58	-
DIEPAMHR + organic matter	356	21.7	22.0	6.6	8.5	7.57	8.11	102.0	58.0	-
High EC Control @ 13.99 mS/cm + organic matter	12930	21.7	22.7	6.7	8.4	7.47	7.95	1560	80	-
High EC Control @ 18.51 mS/cm + organic matter	16780	21.7	22.4	6.9	8.1	7.63	7.98	2200	84	-
High EC Control @ 25.40 mS/cm + organic matter	22850	21.6	22.3	7.0	8.3	7.68	7.97	2960	94	-
Carquinez Strait, West of Benicia army dock (405)	21840	21.7	22.6	6.3	8.7	7.52	7.81	2880	100	0.002
Grizzly Bay at Dolphin (602)	16870	21.7	23.2	6.7	8.7	7.59	7.90	2040	94	0.003
Sacramento R. across from Sherman Lake (704)	2696	21.7	23.0	6.5	8.5	7.64	8.10	320	78	0.003
Suisun Bay off Chipps Island (508)	6160	21.6	23.1	6.7	8.8	7.56	7.97	960	82	0.002
Montezuma Sl. At Nurse Sl. (609)	12805	21.7	23.1	6.5	8.6	7.62	7.87	1880	98	0.000
Middle of Broad Sl., West end (804)	1890	21.7	23.0	6.5	8.9	7.63	8.16	236	76	0.003
Pacheco Creek Outflow (409)	23140	21.8	23.3	6.8	8.8	7.66	7.89	3600	110	0.000
Suisun Bay, East of middle point (504)	10730	21.7	22.1	6.5	8.4	7.61	7.92	1520	88	0.002
Trip Blank (427)	342	21.6	22.8	6.3	8.8	7.66	8.24	112	64	0.001
DIEPAMHR + 25 ppb PBO	341	21.6	22.8	6.2	8.6	7.64	8.16	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	337	21.6	22.9	6.2	8.5	7.60	8.13	-	-	-
High EC Control @ 13.99 mS/cm + organic matter + 25 ppb PBO	71925	21.7	22.9	7.0	8.8	7.55	7.95	-	-	-
High EC Control @ 18.51 mS/cm + organic matter + 25 ppb PBO	18320	21.7	22.7	6.7	8.8	7.58	7.97	-	-	-
High EC Control @ 25.40 mS/cm + organic matter + 25 ppb PBO	22420	21.7	22.9	6.8	8.3	7.65	7.95	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	22445	21.5	22.9	6.4	8.1	7.50	7.82	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	16845	21.5	23.0	6.8	8.7	7.56	7.90	-	-	-
Sacramento R. across from Sherman Lake (704) + 25 ppb PBO	2654.5	21.6	23.1	6.4	8.4	7.58	8.15	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	6040	21.6	22.9	6.8	8.5	7.55	7.95	-	-	-
Montezuma Sl. At Nurse Sl. (609) + 25 ppb PBO	12710	21.5	22.9	6.6	8.3	7.58	7.83	-	-	-
Middle of Broad Sl., West end (804) + 25 ppb PBO	1855	21.5	22.8	6.6	8.9	7.58	8.06	-	-	-
Pacheco Creek Outflow (409) + 25 ppb PBO	23135	21.6	23.1	6.5	8.0	7.61	7.86	-	-	-
Suisun Bay, East of middle point (504) + 25 ppb PBO	10665	21.5	22.9	6.7	8.4	7.56	7.89	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A98-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 10/18/2007 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 10/16/07 - 10/17/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	100	0.0	100	0.0	NS
DIEPAMHR + organic matter	97	2.8	100	0.0	NS
Sacramento R. @ Hood DWR Station	86	5.0	84	4.7	NS
Sacramento R. at tip of Grand Island (711)	90	4.1	100	0.0	NS
San Joaquin R., West of Oulton Point (812)	98	2.5	100	0.0	NS
San Joaquin R. between Hog and Turner Cuts (910)	100	0.0	100	0.0	NS
Old R. at mouth of Holland Cut (915)	98	2.5	98	2.5	NS
Old R. western arm at railroad bridge (902)	100	0.0	98	2.5	NS
Sacramento R. Deep Water Channel, Light 55	100	0.0	100	0.0	NS
Bottle Blank	100	0.0	-	-	NA

	MSD	PMSD
One-way ANOVA	12.0	12.3
Two-way ANOVA	12.0	12.4

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.046	0.004	0.059	0.005	NS
DIEPAMHR + organic matter	0.056	0.006	0.051	0.005	NS
Sacramento R. @ Hood DWR Station	0.062	0.010	0.056	0.007	NS
Sacramento R. at tip of Grand Island (711)	0.067	0.004	0.065	0.004	NS
San Joaquin R., West of Oulton Point (812)	0.078	0.007	0.065	0.005	NS
San Joaquin R. between Hog and Turner Cuts (910)	0.078	0.004	0.088	0.006	NS
Old R. at mouth of Holland Cut (915)	0.067	0.003	0.088	0.004	NS
Old R. western arm at railroad bridge (902)	0.095	0.005	0.060	0.008	Sig (63%)
Sacramento R. Deep Water Channel, Light 55	0.087	0.007	0.068	0.008	NS
Bottle Blank	0.055	0.007	-	-	NA

	MSD	PMSD
One-way ANOVA	0.030	53.0
Two-way ANOVA	0.032	56.0

- Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$). Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
- NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

Table A98-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 10/16//07- 10/17/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Sacramento R. @ Hood DWR Station	215	17.0	7.30	8.6	3.8	0.370	0.002
Sacramento R. at tip of Grand Island (711)	251	16.4	7.57	9.1	6.2	0.370	0.004
San Joaquin R., West of Oulton Point (812)	832	18.3	7.30	9.2	5.0	0.080	0.000
San Joaquin R. between Hog and Turner Cuts (910)	540	18.4	7.30	8.5	5.9	0.100	0.001
Old R. at mouth of Holland Cut (915)	664	17.9	7.63	9.5	3.8	0.040	0.001
Old R. western arm at railroad bridge (902)	744	17.3	7.76	9.4	4.4	0.030	0.000
Sacramento R. Deep Water Channel, Light 55	354	16.9	7.94	9.4	15.3	0.170	0.004
Bottle Blank	-	-	-	-	0.2	-	-

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Table A98-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 10/18/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 10/16/07 - 10/17/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	358	22.8	23.5	6.6	8.4	7.79	8.03	120	60	-
DIEPAMHR + organic matter	361	23.3	23.5	6.7	8.3	7.74	8.07	120	60	-
Sacramento R. @ Hood DWR Station	152	23.6	23.8	5.6	8.6	7.41	7.59	54	59	0.006
Sacramento R. at tip of Grand Island (711)	168	23.6	24.1	6.2	8.5	7.56	7.79	56	58	0.007
San Joaquin R., West of Oulton Point (812)	780	23.7	24.2	6.7	8.7	7.76	7.91	118	67	0.002
San Joaquin R. between Hog and Turner Cuts (910)	464	23.5	23.7	6.6	8.2	7.74	8.04	112	89	0.002
Old R. at mouth of Holland Cut (915)	579	23.7	24.0	6.6	8.7	7.74	7.87	100	69	0.001
Old R. western arm at railroad bridge (902)	659	23.2	23.7	6.6	8.6	7.82	7.92	106	68	0.001
Sacramento R. Deep Water Channel, Light 55	203	23.6	24.0	6.7	8.9	7.77	7.87	66	69	0.005
Bottle Blank	350	23.2	24.0	6.2	8.8	7.72	8.04	106	58	0.000
DIEPAMHR + 25 ppb PBO	355	23.0	23.7	6.8	8.3	7.73	8.05	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	357	23.2	23.6	6.5	8.4	7.80	8.07	-	-	-
Sacramento R. @ Hood DWR Station + 25 ppb PBO	153	23.5	23.9	5.9	8.6	7.50	7.65	-	-	-
Sacramento R. at tip of Grand Island (711) + 25 ppb PBO	166	23.8	23.9	6.0	8.6	7.58	7.76	-	-	-
San Joaquin R., West of Oulton Point (812) + 25 ppb PBO	781	23.2	23.8	6.5	8.7	7.81	7.90	-	-	-
San Joaquin R. between Hog and Turner Cuts (910) + 25 ppb PBO	469	23.9	24.1	6.5	8.4	7.77	8.01	-	-	-
Old R. at mouth of Holland Cut (915) + 25 ppb PBO	583	23.4	23.8	6.7	8.7	7.81	7.89	-	-	-
Old R. western arm at railroad bridge (902) + 25 ppb PBO	658	23.1	24.4	6.7	8.6	7.81	7.94	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	203	23.8	24.1	6.4	8.8	7.70	7.85	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A99-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 10/19/2007 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 10/18/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	100	0.0	98	2.5	NS
DIEPAMHR + organic matter	98	2.5	100	0.0	NS
High EC Control @ 14.97 mS/cm	100	0.0	95	2.9	NS
High EC Control @ 18.62 mS/cm	95	5.0	93	4.8	NS
High EC Control @ 24.20 mS/cm	87	6.3	80	14.1	NS
Montezuma Sl. At Nurse Sl. (609) ³	98	2.5	100	0.0	NS
Suisun Bay off Chipps Island (508)	100	0.0	97	3.1	NS
Grizzly Bay at Dolphin (602) ⁴	90	4.1	89	6.1	NS
Suisun Bay, East of middle point (504) ³	98	2.5	92	2.6	NS
Middle of Broad Sl., West end (804)	100	0.0	100	0.0	NS
Carquinez Strait, West of Benicia army dock (405) ⁵	81	2.8	78	6.3	NS
Sacramento R. across from Sherman Lake (704)	100	0.0	100	0.0	NS
Trip Blank	98	2.5	-	-	NA
Bottle Blank	95	2.9	-	-	NA

	MSD	PMSD
One-way ANOVA	14.9	15.3
Two-way ANOVA	23.0	23.6

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.054	0.009	0.053	0.010	NS
DIEPAMHR + organic matter	0.067	0.007	0.056	0.004	NS
High EC Control @ 14.97 mS/cm	0.081	0.010	0.063	0.008	NS
High EC Control @ 18.62 mS/cm	0.065	0.007	0.063	0.007	NS
High EC Control @ 24.20 mS/cm	0.059	0.002	0.066	0.014	NS
Montezuma Sl. At Nurse Sl. (609) ³	0.072	0.002	0.077	0.008	NS
Suisun Bay off Chipps Island (508)	0.085	0.011	0.068	0.004	NS
Grizzly Bay at Dolphin (602) ⁴	0.063	0.006	0.068	0.008	NS
Suisun Bay, East of middle point (504) ³	0.066	0.009	0.065	0.008	NS
Middle of Broad Sl., West end (804)	0.079	0.004	0.096	0.003	NS
Carquinez Strait, West of Benicia army dock (405) ⁵	0.048	0.006	0.069	0.009	NS
Sacramento R. across from Sherman Lake (704)	0.099	0.007	0.092	0.004	NS
Trip Blank	0.049	0.006	-	-	NA
Bottle Blank	0.043	0.003	-	-	NA

	MSD	PMSD
One-way ANOVA	0.035	52.1
Two-way ANOVA	0.041	60.2

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$). Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.
3. This high conductivity sample was compared to the High EC control @ 14.97 mS/cm.
4. This high conductivity sample was compared to the High EC control @ 18.62 mS/cm.
5. This high conductivity sample was compared to the High EC control @ 24.20 mS/cm.

Table A99-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 10/18/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Montezuma Sl. At Nurse Sl. (609)	14420	17.1	7.59	9.6	8.6	0.070	0.001
Suisun Bay off Chipps Island (508)	7590	16.9	7.81	10.2	4.2	0.080	0.001
Grizzly Bay at Dolphin (602)	17620	16.7	7.81	10.4	9.0	0.100	0.001
Suisun Bay, East of middle point (504)	13270	16.9	7.77	10.2	5.5	0.110	0.001
Middle of Broad Sl., West end (804)	1824	17.3	7.85	10.1	4.6	0.060	0.001
Carquinez Strait, West of Benicia army dock (405)	22250	16.6	7.81	10.3	7.0	0.490	0.006
Sacramento R. across from Sherman Lake (704)	3440	17.2	7.81	10.1	8.4	0.000	0.000
Trip Blank	327	21.2	8.02	8.5	0.2	0.000	0.000
Bottle Blank	-	-	-	-	0.3	-	-

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Table A99-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 10/19/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 10/18/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	352	22.1	23.8	6.7	8.3	7.61	8.02	120	60	-
DIEPAMHR + organic matter	354	22.1	23.7	7.2	8.4	7.68	8.06	120	60	-
High EC Control @ 14.97 mS/cm	13850	22.1	23.8	7.2	8.9	7.25	7.66	1800	82	-
High EC Control @ 18.62 mS/cm	17255	22.1	22.9	6.9	8.4	7.30	7.65	2280	88	-
High EC Control @ 24.20 mS/cm	22585	22.1	23.3	7.1	8.4	7.03	7.65	2880	94	-
Montezuma Sl. At Nurse Sl. (609)	13890	22.1	24.2	7.7	8.9	7.47	7.81	1680	96	0.001
Suisun Bay off Chipps Island (508)	8185	22.1	24.2	7.2	8.8	7.59	7.71	960	82	0.002
Grizzly Bay at Dolphin (602)	17095	22.1	24.3	6.6	8.8	7.57	7.65	2080	92	0.001
Suisun Bay, East of middle point (504)	12050	22.1	23.5	7.0	8.9	7.63	7.68	1440	86	0.002
Middle of Broad Sl., West end (804)	1763	22.1	23.8	6.8	8.9	7.72	7.87	208	72	0.002
Carquinez Strait, West of Benicia army dock (405)	21665	22.1	23.9	6.8	8.4	7.53	7.65	2720	98	0.006
Sacramento R. across from Sherman Lake (704)	3132	22.0	24.4	7.6	8.9	7.76	7.77	356	74	0.002
Trip Blank	354	22.0	23.5	7.0	8.9	7.65	8.05	112	62	0.000
Bottle Blank	356	22.1	23.5	7.3	8.6	7.70	8.05	1120	64	0.000
DIEPAMHR + 25 ppb PBO	346	22.0	23.5	7.0	8.6	7.71	8.03	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	346	22.1	23.9	6.9	8.3	7.64	8.03	-	-	-
High EC Control @ 14.97 mS/cm + 25 ppb PBO	13840	22.1	23.2	7.1	8.7	7.30	7.66	-	-	-
High EC Control @ 18.62 mS/cm + 25 ppb PBO	17290	22.1	23.4	7.2	8.0	7.32	7.65	-	-	-
High EC Control @ 24.20 mS/cm + 25 ppb PBO	22355	22.1	23.6	6.9	8.3	7.10	7.64	-	-	-
Montezuma Sl. At Nurse Sl. (609) + 25 ppb PBO	13875	22.0	23.3	7.1	8.1	7.47	7.77	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	8155	22.0	23.6	7.3	8.8	7.63	7.68	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	16950	22.0	23.7	7.1	8.5	7.58	7.67	-	-	-
Suisun Bay, East of middle point (504) + 25 ppb PBO	12170	22.1	23.6	7.4	8.7	7.65	7.70	-	-	-
Middle of Broad Sl., West end (804) + 25 ppb PBO	1738	22.1	23.6	7.4	8.9	7.76	7.87	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	21585	22.0	23.6	7.0	8.2	7.60	7.63	-	-	-
Sacramento R. across from Sherman Lake (704) + 25 ppb PBO	3151.5	22.1	23.4	7.2	8.9	7.56	7.80	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A100-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 11/1/2007 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 10/30/07 - 10/31/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	100	0.0	98	2.3	NS
DIEPAMHR + organic matter	100	0.0	98	2.3	NS
Sacramento R. at Hood DWR Station	82	2.5	91	5.4	Sig
Sacramento R. Deep Water Channel, Light 55	90	0.0	100	0.0	Sig
San Joaquin R., West of Oulton Point (812)	100	0.0	100	0.0	NS
Old R, at mouth of Holland Cut (915)	100	0.0	100	0.0	NS
Sacramento R. at tip of Grand Island (711)	100	0.0	98	2.5	NS
Old R., western arm at railroad bridge (902)	100	0.0	100	0.0	NS
Sacramento R. across from Sherman Lake (704)	100	0.0	100	0.0	NS
San Joaquin R, between Hog and Turner Cuts (910)	100	0.0	100	0.0	NS
Bottle Blank	100	0.0	-	-	NA
Trip Blank	100	0.0	-	-	NA

	MSD	PMSD
One-way ANOVA	2.6	2.6
Two-way ANOVA	6.1	6.1

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.038	0.009	0.066	0.003	NS
DIEPAMHR + organic matter	0.057	0.004	0.082	0.002	NS
Sacramento R. at Hood DWR Station	0.049	0.006	0.066	0.003	NS
Sacramento R. Deep Water Channel, Light 55	0.061	0.008	0.077	0.006	NS
San Joaquin R., West of Oulton Point (812)	0.038	0.020	0.078	0.011	NS
Old R, at mouth of Holland Cut (915)	0.060	0.006	0.053	0.012	NS
Sacramento R. at tip of Grand Island (711)	0.084	0.008	0.042	0.002	Sig
Old R., western arm at railroad bridge (902)	0.062	0.009	0.052	0.011	NS
Sacramento R. across from Sherman Lake (704)	0.062	0.003	0.066	0.006	NS
San Joaquin R, between Hog and Turner Cuts (910)	0.063	0.015	0.076	0.006	NS
Bottle Blank	0.055	0.006	-	-	NA
Trip Blank	0.064	0.002	-	-	NA

	MSD	PMSD
One-way ANOVA	0.028	49.3
Two-way ANOVA	0.029	51.6

- Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$). Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
- NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

Table A100-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 10/30/07 - 10/31/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Sacramento R. at Hood DWR Station	207	18.0	7.11	9.0	7.5	0.440	0.002
Sacramento R. Deep Water Channel, Light 55	247	16.6	7.59	9.5	15.2	0.200	0.002
San Joaquin R., West of Oulton Point (812)	828	16.8	7.84	9.6	4.9	0.070	0.001
Old R, at mouth of Holland Cut (915)	631	17.1	7.70	9.5	3.9	0.070	0.001
Sacramento R. at tip of Grand Island (711)	250	16.5	7.55	9.4	7.0	0.290	0.003
Old R., western arm at railroad bridge (902)	754	17.0	7.39	9.6	2.6	0.050	0.000
Sacramento R. across from Sherman Lake (704)	3816	16.8	7.89	9.7	17.1	0.110	0.002
San Joaquin R, between Hog and Turner Cuts (910)	633	17.0	7.71	9.6	3.9	0.080	0.001
Bottle Blank	-	-	-	-	0.3	0.010	-
Trip Blank	333	10.2	8.04	9.0	0.3	0.010	0.000

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Table A100-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 11/01/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 10/30/07 - 10/31/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	349	21.5	24.1	7.3	8.6	7.79	8.12	104	59	-
DIEPAMHR + organic matter	348	21.6	23.7	7.3	8.4	7.71	8.06	104	59	-
Sacramento R. at Hood DWR Station	165	21.8	24.5	7.0	8.6	7.54	7.82	56	64	0.015
Sacramento R. Deep Water Channel, Light 55	187	21.5	23.7	6.9	8.4	7.59	8.01	70	74	0.009
San Joaquin R., West of Oulton Point (812)	718	21.6	23.6	7.4	8.5	7.77	7.97	110	68	0.003
Old R, at mouth of Holland Cut (915)	563	21.2	23.4	7.5	8.4	7.80	7.98	98	74	0.003
Sacramento R. at tip of Grand Island (711)	207	21.3	24.5	7.3	8.8	7.60	7.94	66	68	0.012
Old R., western arm at railroad bridge (902)	706	21.0	23.2	7.6	8.4	7.78	8.01	114	71	0.002
Sacramento R. across from Sherman Lake (704)	3752	21.1	24.4	7.5	8.3	7.73	7.92	422	75	0.004
San Joaquin R, between Hog and Turner Cuts (910)	562	20.6	24.3	7.5	8.7	7.90	8.06	134	104	0.003
Bottle Blank	330	21.0	23.6	7.7	8.4	7.80	8.09	106	61	0.001
Trip Blank	348	21.0	24.3	7.7	8.7	7.86	8.07	104	62	0.001
DIEPAMHR + 25 ppb PBO	349	21.4	23.0	7.6	8.3	7.80	8.06	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	352	21.6	23.2	7.5	8.3	7.78	8.08	-	-	-
Sacramento R. at Hood DWR Station + 25 ppb PBO	161	21.2	23.4	7.0	8.2	7.53	7.94	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	187	21.6	23.9	7.6	8.5	7.65	7.97	-	-	-
San Joaquin R., West of Oulton Point (812) + 25 ppb PBO	707	21.0	23.1	7.6	8.3	7.75	7.99	-	-	-
Old R, at mouth of Holland Cut (915) + 25 ppb PBO	558	21.1	23.5	7.7	8.5	7.79	8.01	-	-	-
Sacramento R. at tip of Grand Island (711) + 25 ppb PBO	203	21.0	23.9	7.4	8.6	7.59	7.86	-	-	-
Old R., western arm at railroad bridge (902) + 25 ppb PBO	702	21.4	23.8	7.7	8.5	7.80	8.01	-	-	-
Sacramento R. across from Sherman Lake (704) + 25 ppb PBO	3772	21.5	23.7	7.7	8.4	7.73	7.86	-	-	-
San Joaquin R, between Hog and Turner Cuts (910) + 25 ppb PBO	577	21.5	23.7	7.7	8.7	8.01	8.03	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A101-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 11/2/2007 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 11/01/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	97	2.8	98	2.5	NS
DIEPAMHR + organic matter	98	2.5	95	5.0	NS
High EC Control @ 10.21 mS/cm	100	0.0	98	2.3	NS
High EC Control @ 15.65 mS/cm	100	0.0	100	0.0	NS
High EC Control @ 19.7 mS/cm	100	0.0	98	2.3	NS
High EC Control @ 25.31 mS/cm	89	0.6	93	2.5	NS
Carquinez Strait, West of Benicia army dock (405) ⁶	75	6.8	81	8.3	NS
Grizzly Bay at Dolphin (602) ⁵	88	4.8	95	2.9	NS
Suisun Bay off Chipps Island (508) ³	98	2.5	100	0.0	NS
Middle of Broad Slough, West end (804)	100	0.0	100	0.0	NS
Montezuma Slough at Nurse Slough (609) ⁴	100	0.0	98	2.5	NS
Suisun Bay, East of middle point (504) ⁵	100	0.0	98	2.5	NS

	MSD	PMSD
One-way ANOVA	9.9	10.1
Two-way ANOVA	11.8	12.1

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.038	0.007	0.034	0.006	NS
DIEPAMHR + organic matter	0.031	0.009	0.055	0.007	NS
High EC Control @ 10.21 mS/cm	0.068	0.005	0.073	0.009	NS
High EC Control @ 15.65 mS/cm	0.066	0.008	0.078	0.014	NS
High EC Control @ 19.7 mS/cm	0.041	0.003	0.040	0.004	NS
High EC Control @ 25.31 mS/cm	0.019	0.001	0.046	0.016	NS
Carquinez Strait, West of Benicia army dock (405) ⁶	0.018	0.005	0.028	0.002	NS
Grizzly Bay at Dolphin (602) ⁵	0.044	0.003	0.049	0.007	NS
Suisun Bay off Chipps Island (508) ³	0.052	0.008	0.057	0.010	NS
Middle of Broad Slough, West end (804)	0.070	0.004	0.057	0.005	NS
Montezuma Slough at Nurse Slough (609) ⁴	0.059	0.004	0.055	0.003	NS
Suisun Bay, East of middle point (504) ⁵	0.065	0.005	0.038	0.005	NS

	MSD	PMSD
One-way ANOVA	0.020	63.9
Two-way ANOVA	0.027	87.1

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$). Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.
3. This high conductivity sample was compared to the High EC control @ 10.21 mS/cm.
4. This high conductivity sample was compared to the High EC control @ 15.65 mS/cm.
5. This high conductivity sample was compared to the High EC control @ 19.7 mS/cm.
6. This high conductivity sample was compared to the High EC control @ 25.31 mS/cm.

Table A101-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 11/01/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Carquinez Strait, West of Benicia army dock (405)	23860	16.4	7.83	10.0	9.5	0.120	0.002
Grizzly Bay at Dolphin (602)	18760	16.3	7.8	10.1	25.9	0.120	0.002
Suisun Bay off Chipps Island (508)	9250	16.7	7.75	10.1	9.8	0.080	0.001
Middle of Broad Slough, West end (804)	2833	16.9	7.82	10.0	6.8	0.060	0.001
Montezuma Slough at Nurse Slough (609)	14840	17.2	7.46	8.9	20.4	0.060	0.000
Suisun Bay, East of middle point (504)	16720	16.6	7.79	10.0	12.6	0.100	0.001

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Table A101-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 11/02/2007 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 11/01/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	359	21.1	24.0	7.4	8.4	7.76	8.06	104	59	-
DIEPAMHR + organic matter	358	20.3	24.5	7.8	8.4	7.72	8.01	104	59	-
High EC Control @ 10.21 mS/cm	9785	21.7	23.7	7.2	8.6	7.54	7.85	1120	60	-
High EC Control @ 15.65 mS/cm	14705	20.5	24.3	7.1	8.4	7.52	7.87	1760	80	-
High EC Control @ 19.7 mS/cm	19040	21.1	24.5	7.0	8.5	7.61	7.88	2120	100	-
High EC Control @ 25.31 mS/cm	24420	21.8	24.5	7.2	8.6	7.67	7.89	2800	100	-
Carquinez Strait, West of Benicia army dock (405)	22770	21.5	22.9	6.9	8.4	7.61	7.74	3070	160	0.002
Grizzly Bay at Dolphin (602)	18660	21.5	22.9	7.1	8.8	7.64	7.78	2280	140	0.002
Suisun Bay off Chipps Island (508)	9595	21.9	23.7	7.6	8.6	7.69	7.79	1080	100	0.002
Middle of Broad Slough, West end (804)	2924	22.0	24.3	6.4	8.8	7.75	7.82	332	76	0.002
Montezuma Slough at Nurse Slough (609)	14985	21.6	24.1	7.2	8.5	7.49	7.81	1760	140	0.001
Suisun Bay, East of middle point (504)	17290	22.0	24.5	7.1	8.5	7.66	7.79	1880	140	0.002
DIEPAMHR + 25 ppb PBO	366	22.0	24.2	6.5	8.4	7.81	8.09	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	377	21.9	24.4	6.2	8.8	7.75	8.05	-	-	-
High EC Control @ 10.21 mS/cm + 25 ppb PBO	9965	22.0	24.5	7.5	8.5	7.54	7.84	-	-	-
High EC Control @ 15.65 mS/cm + 25 ppb PBO	15070	22.3	24.5	6.9	8.1	7.56	7.86	-	-	-
High EC Control @ 19.7 mS/cm + 25 ppb PBO	19030	22.4	24.5	7.2	8.3	7.57	7.87	-	-	-
High EC Control @ 25.31 mS/cm + 25 ppb PBO	24220	21.6	24.5	7.0	7.8	7.64	7.88	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	23630	22.7	24.4	7.0	8.0	7.65	7.77	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	18920	22.3	24.5	7.0	8.4	7.62	7.76	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	9575	21.4	24.5	7.2	8.9	7.68	7.78	-	-	-
Middle of Broad Slough, West end (804) + 25 ppb PBO	2913	22.7	24.4	7.1	8.9	7.78	7.84	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	14625	22.2	24.5	7.1	8.3	7.50	7.76	-	-	-
Suisun Bay, East of middle point (504) + 25 ppb PBO	17165	22.7	24.3	6.9	8.6	7.62	7.81	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A102-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 11/15/2007 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 11/13/07 - 11/14/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	98	2.5	98	2.5	NS
DIEPAMHR + organic matter	98	2.5	97	2.8	NS
Sacramento R. at Hood DWR Station	76	3.2	68	4.8	NS
Sacramento R. Deep Water Channel, Light 55	95	2.9	98	2.5	NS
Sacramento R. across from Sherman Lake (704)	100	0.0	98	2.5	NS
Sacramento R. at tip of Grand Island (711)	93	2.5	95	2.9	NS
Bottle Blank	95	5.0	-	-	NA

	MSD	PMSD
One-way ANOVA	13.8	14.1
Two-way ANOVA	13.9	14.3

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.051	0.003	0.048	0.003	NS
DIEPAMHR + organic matter	0.060	0.003	0.055	0.005	NS
Sacramento R. at Hood DWR Station	0.046	0.006	0.057	0.002	NS
Sacramento R. Deep Water Channel, Light 55	0.048	0.003	0.055	0.006	NS
Sacramento R. across from Sherman Lake (704)	0.058	0.009	0.069	0.010	NS
Sacramento R. at tip of Grand Island (711)	0.053	0.007	0.072	0.014	NS
Bottle Blank	0.059	0.008	-	-	NA

	MSD	PMSD
One-way ANOVA	0.028	45.9
Two-way ANOVA	0.033	55.5

- Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$). Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
- NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

Table A102-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 7/25//07- 7/26/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Sacramento R. at Hood DWR Station	235	16.4	7.19	9.1	4.6	0.34	0.001
Sacramento R. Deep Water Channel, Light 55	2984	16.4	7.26	9.6	19.7	0.24	0.001
Sacramento R. across from Sherman Lake (704)	2714	16.5	7.34	9.8	18.2	0.13	0.001
Sacramento R. at tip of Grand Island (711)	269	16.9	7.15	9.2	6.5	0.31	0.001
Bottle Blank	-	-	-	-	0.3	0.00	-

Table A102-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 11/15/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 11/13/07 - 11/14/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	376	21.7	24.9	7.6	8.6	7.84	8.03	104	60	-
DIEPAMHR + organic matter	381	21.7	25.2	7.9	8.4	7.79	8.05	104	60	-
Sacramento R. at Hood DWR Station	203	21.7	25.3	7.3	8.6	7.69	7.75	72	76	0.010
Sacramento R. Deep Water Channel, Light 55	224	21.6	24.7	7.3	8.6	7.78	7.92	72	74	0.010
Sacramento R. across from Sherman Lake (704)	2838	21.7	24.1	7.9	8.6	7.70	7.84	308	70	0.004
Sacramento R. at tip of Grand Island (711)	205	21.7	24.7	7.7	8.7	7.87	7.94	70	70	0.012
Bottle Blank	374	21.7	24.4	7.9	8.6	7.88	8.02	108	57	0.000
DIEPAMHR + 25 ppb PBO	368	21.0	24.2	7.9	8.4	7.88	8.08	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	375	21.4	24.9	7.9	8.6	7.78	8.06	-	-	-
Sacramento R. at Hood DWR Station + 25 ppb PBO	200	21.9	24.6	7.3	8.6	7.58	7.75	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	226	21.9	24.5	7.6	8.5	7.84	7.87	-	-	-
Sacramento R. across from Sherman Lake (704) + 25 ppb PBO	2921	21.7	24.4	7.8	8.4	7.72	7.87	-	-	-
Sacramento R. at tip of Grand Island (711) + 25 ppb PBO	215	21.8	24.4	7.6	8.6	7.85	7.92	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A103-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 11/16/2007 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 11/15/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	100	0.0	92	2.6	NS
DIEPAMHR + organic matter	100	0.0	98	2.5	NS
High EC Control @ 17.58 mS/cm	20	9.1	25	8.7	NS
High EC Control @ 29.52 mS/cm	18	6.3	8	7.5	NS
Old River at mouth of Holland Cut (915)	98	2.5	95	5.0	NS
Suisun Bay, off Chipps Island (508)	90	4.1	97	2.8	NS
Grizzly Bay at Dolphin (602) ³	77	5.3	78	7.9	NS
Montezuma Slough at Nurse Slough (609) ³	95	3.1	100	0.0	NS
San Joaquin, between Hog/Turner Cut (910)	93	4.8	95	2.9	NS
Old River, western arm at railroad bridge (902)	100	0.0	95	2.9	NS
San Joaquin, just west of Oulton Point (812)	100	0.0	95	2.9	NS
Middle of Broad Slough, west end (804)	100	0.0	90	7.1	NS
Suisun Bay, east of middle point (504) ³	89	6.1	95	2.9	NS
Carquinez Straight, just west of Benicia army dock (405) ⁴	21	4.3	12	5.1	NS
Trip Blank	95	2.9	-	-	NA

	MSD	PMSD
One-way ANOVA	21.6	21.6
Two-way ANOVA	21.5	21.5

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.039	0.006	0.046	0.003	NS
DIEPAMHR + organic matter	0.043	0.005	0.070	0.006	NS
High EC Control @ 17.58 mS/cm	0.037	0.018	0.041	0.002	NS
High EC Control @ 29.52 mS/cm	0.040	0.010	0.017	-	NA
Old River at mouth of Holland Cut (915)	0.056	0.004	0.050	0.003	NS
Suisun Bay, off Chipps Island (508)	0.034	0.004	0.038	0.006	NS
Grizzly Bay at Dolphin (602) ³	0.026	0.008	0.027	0.002	NS
Montezuma Slough at Nurse Slough (609) ³	0.039	0.004	0.050	0.005	NS
San Joaquin, between Hog/Turner Cut (910)	0.041	0.006	0.064	0.001	NS
Old River, western arm at railroad bridge (902)	0.050	0.001	0.056	0.004	NS
San Joaquin, just west of Oulton Point (812)	0.056	0.004	0.055	0.004	NS
Middle of Broad Slough, west end (804)	0.050	0.004	0.053	0.004	NS
Suisun Bay, east of middle point (504) ³	0.049	0.008	0.032	0.002	NS
Carquinez Straight, just west of Benicia army dock (405) ⁴	0.034	0.014	0.073	0.012	NS
Trip Blank (427)	0.040	0.002	-	-	NA

	MSD	PMSD
One-way ANOVA	0.036	83.0
Two-way ANOVA	0.033	78.0

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$). Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.
3. This high conductivity sample was compared to the High EC control @ 17.58 mS/cm.
4. This high conductivity sample was compared to the High EC control @ 29.52 mS/cm.

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Table A103-2. Summary of water chemistry measurements on samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 11/15//07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Old River at mouth of Holland Cut (915)	627	17.3	7.11	9.9	3.4	0.04	0.000
Suisun Bay, off Chipps Island (508)	9120	18.9	7.01	9.9	5.8	0.12	0.000
Grizzly Bay at Dolphin (602)	16000	18.4	7.12	9.6	7.0	0.12	0.000
Montezuma Slough at Nurse Slough (609)	15130	18.5	6.89	9.4	18.3	0.08	0.000
San Joaquin, between Hog/Turner Cut (910)	487	17.5	7.08	9.4	3.0	0.04	0.000
Old River, western arm at railroad bridge (902)	644	17.3	7.10	9.8	2.2	0.04	0.000
San Joaquin, just west of Oulton Point (812)	628	16.8	7.18	9.9	5.5	0.10	0.000
Middle of Broad Slough, west end (804)	4316	17.9	7.06	9.9	5.2	0.08	0.000
Suisun Bay, east of middle point (504)	15040	18.4	6.98	10.1	6.1	0.12	0.000
Carquinez Straight, just west of Benicia army dock (405)	28200	17.3	7.29	9.9	6.1	0.13	0.001
Trip Blank (427)	328	20.2	7.69	8.9	0.2	0.00	0.000

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Table A103-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 11/16/2007 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 11/15/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	388	21.8	24.1	6.9	8.5	7.76	8.03	104	60	-
DIEPAMHR + organic matter	387	21.8	24.3	6.8	8.6	7.81	8.01	104	60	-
High EC Control @ 17.58 mS/cm	17760	21.8	23.8	7.6	8.8	7.52	7.88	1960	82	-
High EC Control @ 29.52 mS/cm	29610	22.0	23.8	7.3	8.4	7.56	7.81	3440	104	-
Old River at mouth of Holland Cut (915)	606	22.0	23.8	7.7	8.9	7.78	7.96	98	70	0.001
Suisun Bay, off Chipps Island (508)	9710	21.5	22.8	7.9	8.9	7.64	7.74	1100	83	0.002
Grizzly Bay at Dolphin (602)	17125	21.7	23.7	7.5	8.3	7.55	7.73	1900	91	0.001
Montezuma Slough at Nurse Slough (609)	15240	21.8	23.2	7.4	8.7	7.33	7.71	1740	102	0.001
San Joaquin, between Hog/Turner Cut (910)	448	21.8	23.5	7.0	8.6	7.68	7.94	100	79	0.001
Old River, western arm at railroad bridge (902)	672	21.9	24.0	6.9	8.2	7.73	7.88	104	70	0.001
San Joaquin, just west of Oulton Point (812)	679	22.0	24.1	6.8	8.9	7.70	7.85	104	69	0.002
Middle of Broad Slough, west end (804)	3973.5	21.8	24.0	7.9	8.8	7.62	7.75	422	75	0.001
Suisun Bay, east of middle point (504)	14490	21.8	23.4	7.5	8.7	7.50	7.79	1720	90	0.001
Carquinez Straight, just west of Benicia army dock (405)	28260	21.9	23.8	6.9	8.4	7.51	7.72	3240	104	0.001
Trip Blank (427)	364	21.8	23.2	6.8	8.8	7.77	8.00	110	60	0.000
DIEPAMHR+ 25 ppb PBO	372	21.5	23.7	7.8	8.6	7.82	7.94	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	365	21.7	23.8	7.9	8.7	7.86	8.00	-	-	-
High EC Control @ 17.58 mS/cm + 25 ppb PBO	17230	21.5	24.0	7.7	8.5	7.61	7.83	-	-	-
High EC Control @ 29.52 mS/cm + 25 ppb PBO	28845	21.6	23.8	7.5	8.2	7.62	7.85	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	566.5	21.8	23.7	7.9	8.8	7.78	7.92	-	-	-
Suisun Bay, off Chipps Island (508) + 25 ppb PBO	9815	21.5	23.9	7.8	8.6	7.58	7.73	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	16945	21.6	23.9	7.4	8.5	7.58	7.78	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	15310	21.7	24.0	7.4	8.3	7.43	7.74	-	-	-
San Joaquin, between Hog/Turner Cut (910) + 25 ppb PBO	462	21.6	23.8	7.4	8.7	7.74	7.96	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	667.5	21.7	23.9	7.9	8.9	7.77	7.86	-	-	-
San Joaquin, just west of Oulton Point (812) + 25 ppb PBO	670.5	21.7	23.8	7.7	8.7	7.71	7.85	-	-	-
Middle of Broad Slough, west end (804) + 25 ppb PBO	3985	21.6	23.7	8.0	8.9	7.66	7.85	-	-	-
Suisun Bay, east of middle point (504) + 25 ppb PBO	15240	21.6	23.8	7.5	8.8	7.57	7.78	-	-	-
Carquinez Straight, just west of Benicia army dock (405) + 25 ppb PBO	27915	21.5	23.9	7.3	8.2	7.59	7.75	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A104-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 11/29/07 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 11/27/07 - 11/28/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	98	2.5	98	2.5	NS
DIEPAMHR + organic matter	97	2.8	98	2.5	NS
Sacramento R. at Hood DWR Station	90	7.1	100	0.0	NS
Old R., Western arm at railroad bridge (902)	98	2.5	98	2.5	NS
Sacramento R., across from Sherman Lake (704)	100	0.0	98	2.5	NS
Sacramento R. at tip of Grand Island (711)	85	2.7	83	2.8	NS
Sacramento R. Deep Water Channel, Light 55	82	2.5	75	6.5	NS
San Joaquin R. between Hog and Turner Cuts (910)	98	2.5	100	0.0	NS
Old R. at mouth of Holland Cut (915)	98	2.5	100	0.0	NS
San Joaquin R., West of Oulton Point (812)	100	0.0	100	0.0	NS
Trip Blank	89	0.3	-	-	NA
Bottle Blank	98	2.5	-	-	NA

	MSD	PMSD
One-way ANOVA	14.5	14.9
Two-way ANOVA	15.3	15.8

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.055	0.007	0.053	0.001	NS
DIEPAMHR + organic matter	0.074	0.002	0.067	0.004	NS
Sacramento R. at Hood DWR Station	0.066	0.006	0.060	0.003	NS
Old R., Western arm at railroad bridge (902)	0.080	0.007	0.076	0.003	NS
Sacramento R., across from Sherman Lake (704)	0.064	0.005	0.067	0.006	NS
Sacramento R. at tip of Grand Island (711)	0.058	0.003	0.058	0.004	NS
Sacramento R. Deep Water Channel, Light 55	0.056	0.010	0.067	0.010	NS
San Joaquin R. between Hog and Turner Cuts (910)	0.076	0.007	0.073	0.002	NS
Old R. at mouth of Holland Cut (915)	0.069	0.003	0.072	0.005	NS
San Joaquin R., West of Oulton Point (812)	0.064	0.007	0.072	0.004	NS
Trip Blank	0.059	0.008	-	-	NA
Bottle Blank	0.055	0.003	-	-	NA

	MSD	PMSD
One-way ANOVA	0.031	41.2
Two-way ANOVA	0.029	39.5

- Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$). Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
- NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

Table A104-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 11/27/07 - 11/28/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Sacramento R. at Hood DWR Station	262	13.0	7.16	10.8	4.6	0.37	0.001
Old R., Western arm at railroad bridge (902)	459	14.0	6.91	10.7	3.4	0.10	0.000
Sacramento R., across from Sherman Lake (704)	5050	14.0	7.11	10.6	18.2	0.15	0.000
Sacramento R. at tip of Grand Island (711)	289	13.0	6.90	11.1	6.5	0.25	0.000
Sacramento R. Deep Water Channel, Light 55	336	13.0	6.87	11.3	19.7	0.23	0.000
San Joaquin R. between Hog and Turner Cuts (910)	474	14.2	6.87	10.2	3.4	0.08	0.000
Old R. at mouth of Holland Cut (915)	513	14.1	6.87	10.7	3.4	0.09	0.000
San Joaquin R., West of Oulton Point (812)	743	14.1	6.87	10.6	5.5	0.10	0.000
Trip Blank	348	16.9	8.18	9.5	0.2	0.01	0.000
Bottle Blank	-	-	-	-	0.3	0.00	-

Table A104-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 11/29/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 11/27/07 - 11/28/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	359	21.3	22.4	7.6	8.6	7.74	8.03	112	62	-
DIEPAMHR + organic matter	360	21.9	22.2	7.8	8.8	7.68	8.01	112	62	-
Sacramento R. at Hood DWR Station	214	22.1	23.4	7.0	8.4	7.53	7.88	84	80	0.013
Old R., Western arm at railroad bridge (902)	571	21.8	22.6	7.5	8.2	7.69	7.96	272	76	0.004
Sacramento R., across from Sherman Lake (704)	4960	21.9	22.1	7.6	8.9	7.65	7.78	560	83	0.003
Sacramento R. at tip of Grand Island (711)	226	21.9	22.8	7.4	8.6	7.70	7.99	74	82	0.011
Sacramento R. Deep Water Channel, Light 55	237	21.7	23.0	7.2	8.0	7.74	7.91	82	88	0.008
San Joaquin R. between Hog and Turner Cuts (910)	382	21.8	22.8	7.5	8.1	7.75	8.01	100	81	0.003
Old R. at mouth of Holland Cut (915)	467	22.5	22.7	7.5	8.3	7.75	7.98	96	73	0.004
San Joaquin R., West of Oulton Point (812)	707	22.2	23.2	7.4	8.3	7.72	7.94	112	76	0.004
Trip Blank	357	22.1	22.6	7.6	8.2	7.73	8.05	108	61	0.000
Bottle Blank	360	22.1	22.7	7.7	8.3	7.74	8.04	104	61	0.000
DIEPAMHR + 25 ppb PBO	351	21.8	22.0	7.7	8.4	7.68	8.05	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	354	21.8	22.2	7.6	8.6	7.67	8.02	-	-	-
Sacramento R. at Hood DWR Station + 25 ppb PBO	210	21.8	22.3	6.8	8.3	7.52	7.98	-	-	-
Old R., Western arm at railroad bridge (902) + 25 ppb PBO	556	21.5	22.0	7.5	8.3	7.75	7.99	-	-	-
Sacramento R., across from Sherman Lake (704) + 25 ppb PBO	5060	21.8	22.0	7.6	8.1	7.67	7.81	-	-	-
Sacramento R. at tip of Grand Island (711) + 25 ppb PBO	215	22.1	22.2	7.5	8.2	7.75	8.15	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	227	21.9	22.1	7.5	8.7	7.75	8.00	-	-	-
San Joaquin R. between Hog and Turner Cuts (910) + 25 ppb PBO	374	21.8	21.9	7.5	8.3	7.76	8.02	-	-	-
Old R. at mouth of Holland Cut (915) + 25 ppb PBO	462	21.9	21.9	7.6	8.3	7.79	8.01	-	-	-
San Joaquin R., West of Oulton Point (812) + 25 ppb PBO	693	21.7	21.7	7.4	8.5	7.70	7.95	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A105-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 11/30/07 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 11/29/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	90	0.0	98	2.5	NS
DIEPAMHR + organic matter	88	4.8	100	0.0	NS
High EC Control @ 12.59 mS/cm	100	0.0	100	0.0	NS
High EC Control @ 18.56 mS/cm	93	4.8	98	2.5	NS
Grizzly Bay at Dolphin (602)	95	2.9	98	2.5	NS
Middle of Broad Slough, West end (804)	98	2.5	100	0.0	NS
Suisun Bay, East of middle point (504) ⁴	95	3.1	100	0.0	NS
Montezuma Slough at Nurse Slough (609) ⁴	100	0.0	100	0.0	NS
Suisun Bay off Chipps Island (508) ³	100	0.0	97	2.8	NS

	MSD	PMSD
One-way ANOVA	13.4	15.3
Two-way ANOVA	12.0	13.7

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.043	0.005	0.049	0.004	NS
DIEPAMHR + organic matter	0.049	0.003	0.055	0.005	NS
High EC Control @ 12.59 mS/cm	0.086	0.002	0.035	0.003	S (41%)
High EC Control @ 18.56 mS/cm	0.024	0.003	0.026	0.002	NS
Grizzly Bay at Dolphin (602)	0.038	0.004	0.044	0.006	NS
Middle of Broad Slough, West end (804)	0.072	0.006	0.067	0.016	NS
Suisun Bay, East of middle point (504) ⁴	0.021	0.006	0.032	0.003	NS
Montezuma Slough at Nurse Slough (609) ⁴	0.039	0.009	0.035	0.006	NS
Suisun Bay off Chipps Island (508) ^{3,5}	0.032	0.001	0.046	0.006	NS

	MSD	PMSD
One-way ANOVA	0.024	48.8
Two-way ANOVA	0.031	63.1

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.

Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. This high conductivity sample was compared to the High EC control @ 12.59 mS/cm.

4. This high conductivity sample was compared to the High EC control @ 18.56 mS/cm.

5. Although sample 508 showed significantly lower weights than the High EC control @ 12.59 mS/cm, this control showed anomalously high weights compared to the other treatments in this test. *Hyaella* weight is not typically enhanced in this conductivity range (SEE FIGURE X), and in our best professional judgement it is more reasonable to use the weights observed in the primary control treatment to evaluate the performance of animals exposed to sample 508.

Table A105-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 11/29/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Grizzly Bay at Dolphin (602)	16640	12.5	7.70	10.9	11.0	0.15	0.001
Middle of Broad Slough, West end (804)	4718	13.3	7.38	10.8	8.8	0.12	0.001
Suisun Bay, East of middle point (504)	16940	12.9	7.57	10.9	14.2	0.16	0.001
Montezuma Slough at Nurse Slough (609)	14940	12.4	7.21	9.9	27.4	0.27	0.001
Suisun Bay off Chipps Island (508)	12250	12.8	7.54	11.0	14.9	0.15	0.001

Table A105-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 11/30/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 11/29/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	360	22.1	22.5	7.7	8.7	7.76	7.99	112	62	-
DIEPAMHR + organic matter	360	22.4	23.0	7.5	8.6	7.72	8.00	112	62	-
High EC Control @ 12.59 mS/cm	12520	22.4	22.8	7.5	8.2	7.68	7.94	1380	78	-
High EC Control @ 18.56 mS/cm	18185	22.5	22.8	7.1	8.4	7.62	7.91	2120	88	-
Grizzly Bay at Dolphin (602)	17000	22.4	22.8	7.0	8.1	7.69	7.78	1920	94	0.003
Middle of Broad Slough, West end (804)	4760	22.0	22.1	7.2	8.7	7.77	7.82	560	80	0.003
Suisun Bay, East of middle point (504)	18045	22.3	22.5	7.1	8.3	7.71	7.79	1940	94	0.003
Montezuma Slough at Nurse Slough (609)	15490	21.9	22.7	7.3	8.3	7.58	7.84	1780	102	0.003
Suisun Bay off Chipps Island (508)	12655	21.9	23.2	7.4	8.5	7.74	7.81	1360	86	0.003
DIEPAMHR + 25 ppb PBO	382	22.0	23.1	7.6	8.4	7.76	8.17	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	383	22.2	22.5	7.5	8.6	7.73	8.06	-	-	-
High EC Control @ 12.59 mS/cm + 25 ppb PBO	12565	22.3	23.8	7.5	8.6	7.70	7.91	-	-	-
High EC Control @ 18.56 mS/cm + 25 ppb PBO	16955	22.3	23.2	6.4	8.3	7.57	7.91	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	17065	22.0	23.4	7.2	8.3	7.73	7.83	-	-	-
Middle of Broad Slough, West end (804) + 25 ppb PBO	4803	22.1	23.2	7.6	8.7	7.79	7.87	-	-	-
Suisun Bay, East of middle point (504) + 25 ppb PBO	17815	22.0	23.5	7.2	8.4	7.72	7.82	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	15355	22.0	22.9	7.3	8.5	7.74	7.84	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	12700	22.0	23.0	7.4	8.4	7.76	7.81	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A106-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 12/13/07 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 12/11/07 - 12/12/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	100	0.0	98	2.5	NS
DIEPAMHR + Organic Matter	89	4.5	100	0.0	NS
Sacramento R. @ Hood DWR Station	98	2.5	98	2.5	NS
Old R., western arm at railroad bridge (902)	97	2.8	100	0.0	NS
Old R. at mouth of Holland Cut (915)	100	0.0	100	0.0	NS
San Joaquin R. between Hog and Turner Cuts (910)	100	0.0	98	2.3	NS
San Joaquin R., West of Oulton Point (812)	100	0.0	100	0.0	NS
Sacramento R. Deep Water Channel, Light 55	100	0.0	100	0.0	NS
Sacramento R. at tip of Grand Island (711)	95	2.9	90	4.1	NS
Field Dup.: San Joaquin R. between Hog and Turner Cuts (910)	98	2.5	-	-	NA
Field Dup.: Old R., western arm at railroad bridge (902)	100	0.0	-	-	NA
Field Dup.: Sacramento R. @ Hood DWR Station	98	2.5	-	-	NA
Bottle Blank (227)	100	0.0	-	-	NA
Bottle Blank (228)	98	2.5	-	-	NA
Bottle Blank (229)	98	2.5	-	-	NA
Bottle Blank (230)	98	2.5	-	-	NA

	MSD	PMSD
One-way ANOVA	11.0	12.3
Two-way ANOVA	10.8	12.0

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.047	0.004	0.065	0.008	NS
DIEPAMHR + Organic Matter	0.058	0.005	0.070	0.003	NS
Sacramento R. @ Hood DWR Station	0.066	0.009	0.059	0.005	NS
Old R., western arm at railroad bridge (902)	0.073	0.005	0.081	0.006	NS
Old R. at mouth of Holland Cut (915)	0.079	0.002	0.063	0.003	NS
San Joaquin R. between Hog and Turner Cuts (910)	0.078	0.005	0.067	0.012	NS
San Joaquin R., West of Oulton Point (812)	0.076	0.007	0.067	0.010	NS
Sacramento R. Deep Water Channel, Light 55	0.071	0.009	0.077	0.006	NS
Sacramento R. at tip of Grand Island (711)	0.045	0.012	0.051	0.004	NS
Field Dup.: San Joaquin R. between Hog and Turner Cuts (910)	0.096	0.008	-	-	NA
Field Dup.: Old R., western arm at railroad bridge (902)	0.068	0.006	-	-	NA
Field Dup.: Sacramento R. @ Hood DWR Station	0.058	0.008	-	-	NA
Bottle Blank (227)	0.047	0.007	-	-	NA
Bottle Blank (228)	0.042	0.010	-	-	NA
Bottle Blank (229)	0.050	0.004	-	-	NA
Bottle Blank (230)	0.034	0.009	-	-	NA

	MSD	PMSD
One-way ANOVA	0.038	65.2
Two-way ANOVA	0.039	66.5

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.
Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

Table A106-2. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 12/11/07 - 12/12/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Sacramento R. @ Hood DWR Station	205	10.8	7.03	11.4	12.1	0.30	0.001
Old R., western arm at railroad bridge (902)	696	10.7	7.18	11.3	7.0	0.16	0.000
Old R. at mouth of Holland Cut (915)	660	10.5	7.23	11.2	3.6	0.15	0.000
San Joaquin R. between Hog and Turner Cuts (910)	437	10.9	7.06	10.8	4.4	0.09	0.000
San Joaquin R., West of Oulton Point (812)	802	11.0	7.17	11.2	6.2	0.16	0.000
Sacramento R. Deep Water Channel, Light 55	328	10.7	7.05	11.7	16.6	0.22	0.000
Sacramento R. at tip of Grand Island (711)	272	10.4	7.14	10.7	7.8	0.31	0.001
Field Dup.: San Joaquin R. between Hog and Turner Cuts (910)	437	10.9	7.06	10.8	3.9	0.08	0.000
Field Dup.: Old R., western arm at railroad bridge (902)	696	10.7	7.18	11.3	5.2	0.12	0.000
Field Dup.: Sacramento R. @ Hood DWR Station	205	10.8	7.03	11.4	11.9	0.29	0.001
Bottle Blank (227)	-	-	-	-	0.2	0.00	-
Bottle Blank (228)	-	-	-	-	0.3	0.01	-
Bottle Blank (229)	-	-	-	-	0.2	0.00	-
Bottle Blank (230)	-	-	-	-	0.2	0.02	-

Table A106-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 12/13/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 12/11/07 - 12/12/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	365	22.6	23.2	7.4	8.6	7.75	8.15	100	60	-
DIEPAMHR + Organic Matter	359	22.7	22.9	7.3	8.5	7.70	8.14	100	60	-
Sacramento R. @ Hood DWR Station	217	22.6	22.8	6.8	8.8	7.67	8.09	88	86	0.016
Old R., western arm at railroad bridge (902)	697	22.6	23.4	7.4	8.5	7.74	8.04	100	74	0.008
Old R. at mouth of Holland Cut (915)	689	22.5	23.4	7.4	8.9	7.79	8.05	116	54	0.007
San Joaquin R. between Hog and Turner Cuts (910)	409	22.5	23.4	7.4	8.9	7.83	8.10	88	84	0.005
San Joaquin R., West of Oulton Point (812)	775	22.5	23.2	7.5	8.9	7.73	8.10	124	76	0.008
Sacramento R. Deep Water Channel, Light 55	268	22.4	23.4	7.4	8.6	7.89	8.19	88	90	0.015
Sacramento R. at tip of Grand Island (711)	220	22.4	23.2	7.4	8.9	7.82	8.10	80	80	0.017
Field Dup.: San Joaquin R. between Hog and Turner Cuts (910)	414	22.3	23.6	7.5	8.9	7.81	8.13	104	80	0.005
Field Dup.: Old R., western arm at railroad bridge (902)	679	22.3	23.2	7.4	8.9	7.80	8.05	116	74	0.006
Field Dup.: Sacramento R. @ Hood DWR Station	218	22.3	23.6	6.9	8.8	7.70	8.02	80	86	0.014
Bottle Blank (227)	361	22.5	23.3	7.5	8.8	7.72	8.06	104	48	0.000
Bottle Blank (228)	353	22.1	23.0	7.6	8.9	7.71	8.00	108	78	0.000
Bottle Blank (229)	356	22.1	23.0	7.7	8.6	7.75	8.08	108	68	0.000
Bottle Blank (230)	364	22.0	23.0	7.6	8.7	7.79	8.08	104	58	0.001
DIEPAMHR + 25 ppb PBO	349	22.6	22.9	7.4	8.5	7.71	8.08	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	342	22.5	22.8	7.4	8.5	7.72	8.02	-	-	-
Sacramento R. @ Hood DWR Station + 25 ppb PBO	217	22.5	22.7	6.9	8.9	7.70	8.04	-	-	-
Old R., western arm at railroad bridge (902) + 25 ppb PBO	690	22.4	22.9	7.1	8.6	7.75	8.07	-	-	-
Old R. at mouth of Holland Cut (915) + 25 ppb PBO	640	22.4	22.8	7.4	8.8	7.79	8.05	-	-	-
San Joaquin R. between Hog and Turner Cuts (910) + 25 ppb PBO	419	22.4	22.8	7.2	8.6	7.78	8.07	-	-	-
San Joaquin R., West of Oulton Point (812) + 25 ppb PBO	772	22.4	22.6	7.3	8.7	7.76	7.92	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	264	22.3	23.1	7.2	8.9	7.80	8.12	-	-	-
Sacramento R. at tip of Grand Island (711) + 25 ppb PBO	217	22.3	22.8	7.2	8.5	7.75	7.97	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

Table A107-1. Summary of a 10-day *H. azteca* water column toxicity test initiated on 12/14/07 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 12/13/07.

Treatment	Survival (%) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	100	0.0	98	2.5	NS
DIEPAMHR + organic matter	100	0.0	98	2.5	NS
High EC Control @ 13.39 mS+ organic matter	100	0.0	98	2.5	NS
High EC Control @ 16.54 mS+ organic matter	100	0.0	98	2.5	NS
DIEPAMHR + 75 ng/L permethrin	3	2.5	0	0.0	NS
Suisun Bay, East of middle point (504)	100	0.0	95	2.9	NS
Grizzly Bay at Dolphin (602)	95	2.9	100	0.0	NS
Suisun Bay off Chipps Island (508)	97	2.8	100	0.0	NS
Sacramento R. across from Sherman Lake (704)	100	0.0	100	0.0	NS
Middle of Broad Slough, West end (804)	98	2.5	100	0.0	NS
Montezuma Slough at Nurse Slough (609)	98	2.3	100	0.0	NS
Field Dup.: Montezuma Slough at Nurse Slough (609)	100	0.0	-	-	NA
Bottle Blank (231)	98	2.5	-	-	NA
Bottle Blank (232)	100	0.0	-	-	NA
DIEPAMHR (renewed every 2 days)	100	0.0	100	0.0	NS
DIEPAMHR + 75 ng/L Permethrin (renewed every 2 days)	0	0.0	0	0.0	NS

	MSD	PMSD
One-way ANOVA	8.0	8.0
Two-way ANOVA	9.1	9.1

Treatment	Weight (mg/surviving individual) ¹				
	Unmanipulated		25 ppb PBO added		vs Non-PBO ²
	mean	se	mean	se	
DIEPAMHR	0.065	0.010	0.078	0.006	NS
DIEPAMHR + organic matter	0.098	0.008	0.090	0.008	NS
High EC Control @ 13.39 mS+ organic matter	0.071	0.002	0.068	0.006	NS
High EC Control @ 16.54 mS+ organic matter	0.063	0.004	0.071	0.003	NS
DIEPAMHR + 75 ng/L permethrin ⁵	0.230	-	-	-	NA
Suisun Bay, East of middle point (504)	0.073	0.006	0.066	0.008	NS
Grizzly Bay at Dolphin (602)	0.071	0.004	0.079	0.004	NS
Suisun Bay off Chipps Island (508)	0.078	0.009	0.087	0.003	NS
Sacramento R. across from Sherman Lake (704)	0.092	0.006	0.087	0.005	NS
Middle of Broad Slough, West end (804)	0.096	0.006	0.101	0.007	NS
Montezuma Slough at Nurse Slough (609)	0.095	0.002	0.089	0.006	NS
Field Dup.: Montezuma Slough at Nurse Slough (609)	0.090	0.003	-	-	NA
Bottle Blank (231)	0.085	0.004	-	-	NA
Bottle Blank (232)	0.084	0.003	-	-	NA
DIEPAMHR (renewed every 2 days)	0.066	0.005	0.059	0.003	NS
DIEPAMHR + 75 ng/L Permethrin (renewed every 2 days)	-	-	-	-	NA

	MSD	PMSD
One-way ANOVA	0.029	29.9
Two-way ANOVA	0.032	32.3

- Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Unmanipulated samples were analyzed using one-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$). Samples with PBO additions were analyzed using two-way ANOVA and Tukey's Multiple Comparison Procedure ($P < 0.05$).
- NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.
- This high conductivity sample was compared to the High EC control @ 13.39 mS/cm.
- This high conductivity sample was compared to the High EC control @ 16.54 mS/cm.
- DIEPAMHR + 75 ng/L permethrin exhibited 3% survival, however average weight exceeded all other weights at 0.230 mg/individual. This anomalous weight is due to the error of the scale used to determine weights rather than abnormally large *H. azteca*.

Table A107-2. Daily survival of *H. azteca* exposed to control water spiked with permethrin during a 10-day water column toxicity test initiated on 12/14/07 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 12/13/07.

Treatment	Renewal	Treatment ID	Survival (%)				
			Day 1	Day 2	Day 3	Day 4	Day 10
Control	5 day	131	100	100	100	100	100
Control + 25 ppb PBO	5 day	145	100	98	98	98	98
Control	2 day	156	100	100	100	100	100
Control + 25 ppb PBO	2 day	158	100	100	100	100	100
75 ng/L Permethrin	5 day	135	15	8	5	3	3
75 ng/L Permethrin + 25 ppb PBO	5 day	149	8	5	0	0	0
75 ng/L Permethrin	2 day	157	35	23	3	3	0
75 ng/L Permethrin + 25 ppb PBO	2 day	159	23	10	0	0	0

Table A107-3. Summary of water chemistry at field conditions of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 12/13/07.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Suisun Bay, East of middle point (504)	16920	10.5	7.56	11.8	15.7	0.24	0.001
Grizzly Bay at Dolphin (602)	15660	10.0	7.45	12.1	10.7	0.21	0.001
Suisun Bay off Chipps Island (508)	12200	10.1	7.20	11.9	15.1	0.21	0.000
Sacramento R. across from Sherman Lake (704)	2957	10.0	7.25	11.9	13.1	0.26	0.001
Middle of Broad Slough, West end (804)	4146	10.5	6.80	11.8	14.2	0.23	0.000
Montezuma Slough at Nurse Slough (609)	6840	9.6	7.12	11.9	11.9	0.23	0.000
Field Dup.: Montezuma Slough at Nurse Slough (609)	6840	9.6	7.12	11.9	15.6	0.24	0.000
Bottle Blank (231)	-	-	-	-	0.2	0.02	-
Bottle Blank (232)	-	-	-	-	0.2	0.01	-

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Table A107-4. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 12/14/07 of samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 12/13/07.

Treatment	Laboratory Chemistry							Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Unionized Ammonia (mg/L) ¹
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L) ²	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	352	22.8	22.8	7.4	8.7	7.77	8.41	100	60	-
DIEPAMHR + organic matter	358	22.5	22.8	7.2	8.7	7.71	8.26	100	60	-
High EC Control @13.39 mS + organic matter	12690	22.5	22.7	7.3	8.9	7.67	7.77	-	-	-
High EC Control @16.54 mS + organic matter	16185	22.5	22.5	7.4	8.6	7.67	7.77	-	-	-
DIEPAMHR + 75 ng/L permethrin	755	22.3	22.9	6.5	8.9	7.65	8.28	-	-	-
Suisun Bay, East of middle point (504)	17025	22.5	23.0	7.3	8.8	7.62	7.88	1000	90	0.004
Grizzly Bay at Dolphin (602)	15900	22.4	22.9	7.2	8.8	7.66	7.90	1120	92	0.004
Suisun Bay off Chipps Island (508)	12660	22.3	23.7	7.7	8.6	7.69	7.94	1400	88	0.005
Sacramento R. across from Sherman Lake (704)	3214	22.3	23.7	7.4	8.9	7.77	8.23	332	80	0.010
Middle of Broad Slough, West end (804)	4363	22.3	23.8	7.5	8.8	7.75	8.06	464	82	0.007
Montezuma Slough at Nurse Slough (609)	7035	22.4	23.8	7.5	8.9	7.68	8.03	728	82	0.006
Field Dup.: Montezuma Slough at Nurse Slough (609)	7170	22.1	23.8	7.4	8.9	7.70	8.00	732	84	0.005
Bottle Blank (231)	537	22.3	24.0	7.5	8.8	7.82	8.40	100	58	0.001
Bottle Blank (232)	374	22.2	23.8	7.4	8.6	7.82	8.29	104	58	0.000
DIEPAMHR + 25 ppb PBO	364	22.2	22.5	7.4	8.7	7.66	8.22	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	359	22.5	22.6	7.3	8.6	7.64	8.28	-	-	-
High EC Control @13.39 mS + organic matter + 25 ppb PBO	12730	22.2	22.5	7.4	8.6	7.67	7.80	-	-	-
High EC Control @16.54 mS + organic matter + 25 ppb PBO	16065	22.2	22.6	7.4	8.7	7.69	7.80	-	-	-
DIEPAMHR + 75 ng/L permethrin + 25 ppb PBO	385	22.7	22.7	2.7	8.7	7.55	8.09	-	-	-
Suisun Bay, East of middle point (504) + 25 ppb PBO	17320	22.3	22.5	7.1	8.4	7.66	7.78	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	15805	22.4	22.6	7.0	8.6	7.65	7.83	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	12440	22.4	22.6	7.4	8.7	7.68	7.90	-	-	-
Sacramento R. across from Sherman Lake (704) + 25 ppb PBO	3203	22.4	22.7	7.4	8.9	7.79	8.19	-	-	-
Middle of Broad Slough, West end (804) + 25 ppb PBO	4333	22.5	22.7	7.2	8.8	7.78	8.11	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	7120	22.6	22.6	7.6	8.7	7.74	7.97	-	-	-
DIEPAMHR (renewed every 2 days)	420	22.6	23.6	7.6	8.5	7.77	8.49	100	60	-
DIEPAMHR + 75 ng/L Permethrin (renewed every 2 days)	245	20.6	23.6	4.5	8.9	7.61	7.96	100	60	-
DIEPAMHR (renewed every 2 days) + 25 ppb PBO	368	22.6	22.9	7.6	8.8	7.90	8.37	-	-	-
DIEPAMHR + 75 ng/L Permethrin (renewed every 2 days) + 25 ppb PBO	1799	21.9	22.7	3.6	8.6	7.40	8.01	-	-	-

1: This Unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at test initiation.

2: Highlighted cells indicate anomolous dissolved oxygen measurements. It is unlikely that these low DO measurements were the cause of high mortality exhibited in these samples, as the samples spiked with permethrin without the addition of PBO also exhibited high mortality.

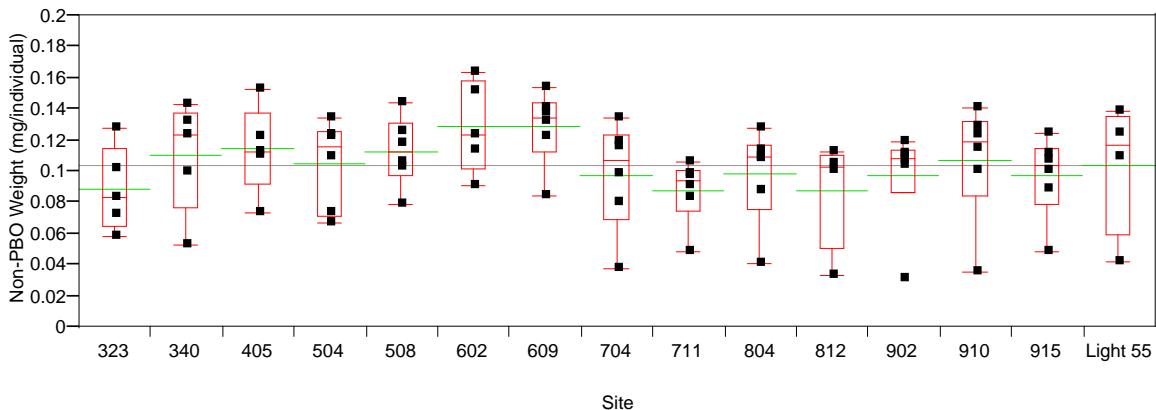
Appendix B

Hyalella azteca

Weight and PBO Effects by Season

Appendix B. Section 1. Weights of *H. azteca* at each site during every season, and tables of model coefficients showing possible between-site effects on weight after controlling for the effects of differences in conductivity. Note that weight as a percentage of the control weight was used as the response variable in the ANCOVA models in order to minimize variation due to differences in the size of *H. azteca* used in different tests.

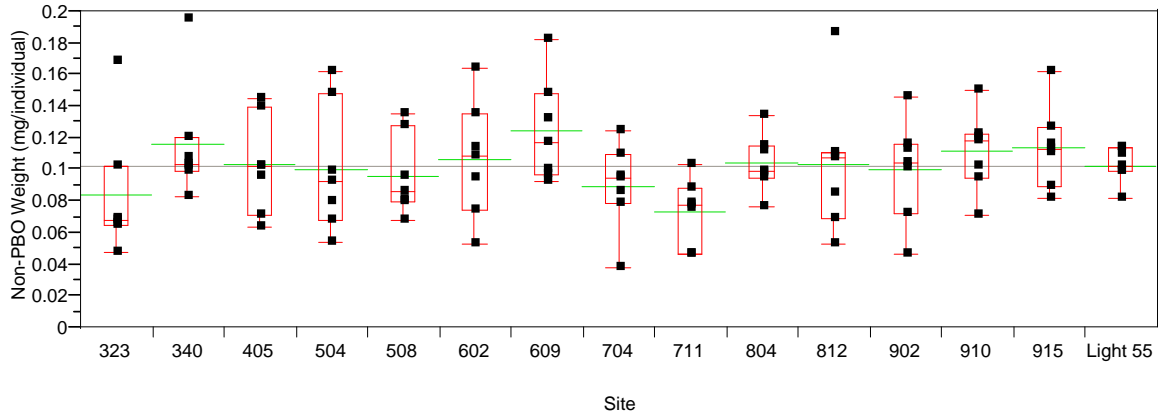
Winter 2006



Effects on Non-PBO Weight (measured as % of Non-PBO Control Weight)

Term	Scaled Estimate	Plot Estimate	Std Error	t Ratio	Prob> t
Intercept	164.8		7.9	20.93	<.0001
Log EC	30.2		22.8	1.32	0.1919
(Log EC) ²	-41.5		20.8	-2.00	0.0505
Sample[323]	-26.3		24.3	-1.08	0.2836
Sample[340]	-4.7		21.8	-0.22	0.8301
Sample[405]	-2.4		18.7	-0.13	0.8981
Sample[504]	-10.7		14.5	-0.74	0.4639
Sample[508]	-1.7		16.1	-0.10	0.9175
Sample[602]	21.7		15.7	1.38	0.1718
Sample[609]	8.2		16.7	0.49	0.6256
Sample[704]	0.2		16.6	0.01	0.9903
Sample[711]	2.5		19.3	0.13	0.8977
Sample[804]	4.8		17.5	0.27	0.7849
Sample[812]	-1.7		19.5	-0.09	0.9317
Sample[902]	-6.8		16.1	-0.42	0.6735
Sample[910]	11.3		16.1	0.70	0.4852
Sample[915]	-3.9		16.1	-0.24	0.8101
Sample[Light 55]	9.5		17.7	0.54	0.5941

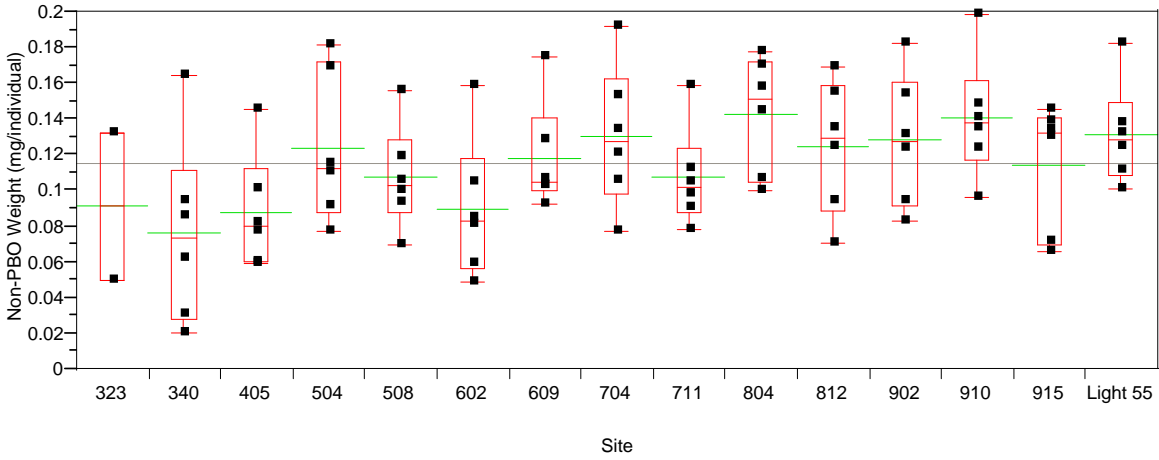
Spring 2006



Effects on Non-PBO Weight (measured as % of Non-PBO Control Weight)

Term	Scaled Estimate	Plot Estimate	Std Error	t Ratio	Prob> t
Intercept	157.0		7.3	21.47	<.0001
Log EC	40.3		31.0	1.30	0.1972
(Log EC) ²	-57.9		26.8	-2.16	0.0334
Sample[323]	-0.1		26.7	-0.00	0.9974
Sample[340]	13.2		21.9	0.60	0.5489
Sample[405]	-7.4		17.8	-0.41	0.6807
Sample[504]	-9.4		15.3	-0.61	0.5415
Sample[508]	-14.8		16.3	-0.91	0.3673
Sample[602]	-2.4		15.0	-0.16	0.8738
Sample[609]	8.3		16.6	0.50	0.6179
Sample[704]	-11.4		15.6	-0.73	0.4683
Sample[711]	-37.4		17.2	-2.18	0.0323
Sample[804]	19.94		16.0	1.24	0.2172
Sample[812]	24.4		16.1	1.52	0.1319
Sample[902]	-9.7		16.3	-0.60	0.5533
Sample[910]	13.8		16.6	0.83	0.4105
Sample[915]	23.3		16.2	1.43	0.1554
Sample[Light 55]	-10.5		17.8	-0.59	0.5557

Summer 2006

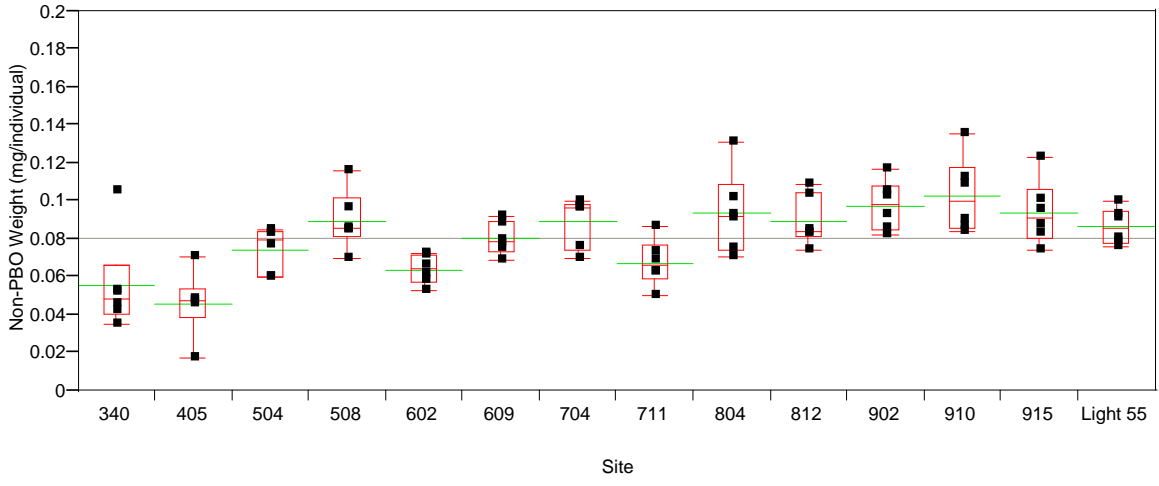


Effects on Non-PBO Weight (measured as % of Non-PBO Control Weight)

Term	Scaled Estimate	Plot Estimate	Std Error	t Ratio	Prob> t
Intercept	155.0		14.1	10.97	<.0001
Log EC	-18.2		28.7	-0.64	0.5270
(Log EC) ²	-47.6		31.0	-1.53	0.1294
Sample[340]	16.8		37.3	0.45	0.6527
Sample[405]	24.7		31.3	0.79	0.4316
Sample[504]	9.4		22.7	0.41	0.6803
Sample[508]	0.0		19.1	0.00	0.9983
Sample[602]	-5.3		26.2	-0.20	0.8397
Sample[609]	18.9		19.2	0.99	0.3263
Sample[704]	-24.1		20.1	-1.20	0.2340
Sample[711]	-13.2		24.3	-0.54	0.5898
Sample[804]	-15.1		17.6	-0.86	0.3932
Sample[812]	-28.0		20.9	-1.34	0.1862
Sample[902]	4.4		21.0	0.21	0.8357
Sample[910]	17.6		17.9	0.99	0.3280
Sample[915]	-13.3		21.0	-0.63	0.5296
Sample[Light 55]	7.0		18.5	0.38	0.7062

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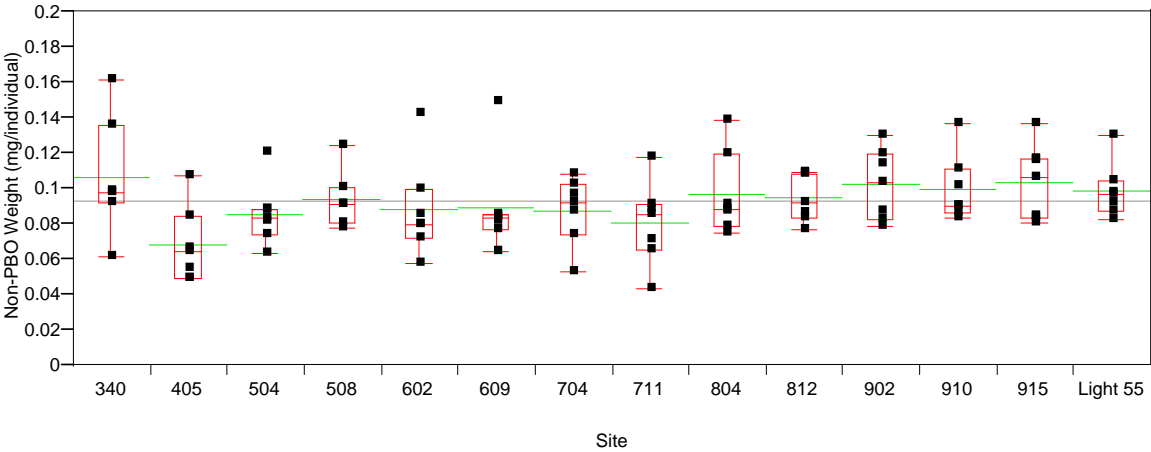


Effects on Non-PBO Weight (measured as % of Non-PBO Control Weight)

Term	Scaled Estimate	Plot Estimate	Std Error	t Ratio	Prob> t
Intercept	137.8		7.8	17.58	<.0001
Log EC	4.4		13.6	0.32	0.7478
(Log EC) ²	-13.6		15.7	-0.87	0.3891
Sample[340]	-33.7		15.5	-2.18	0.0327
Sample[405]	-47.7		16.9	-2.82	0.0062
Sample[504]	-0.3		12.7	-0.02	0.9825
Sample[508]	23.2		11.6	2.00	0.0494
Sample[602]	-18.7		14.4	-1.30	0.1979
Sample[609]	10.5		13.2	0.80	0.4281
Sample[704]	2.5		10.8	0.24	0.8147
Sample[711]	-18.7		16.3	-1.15	0.2538
Sample[804]	7.7		11.2	0.68	0.4966
Sample[812]	8.3		13.1	0.64	0.5268
Sample[902]	18.8		13.0	1.44	0.1539
Sample[910]	28.1		13.1	2.15	0.0353
Sample[915]	13.6		13.3	1.02	0.3097
Sample[Light 55]	6.4		14.5	0.44	0.6602

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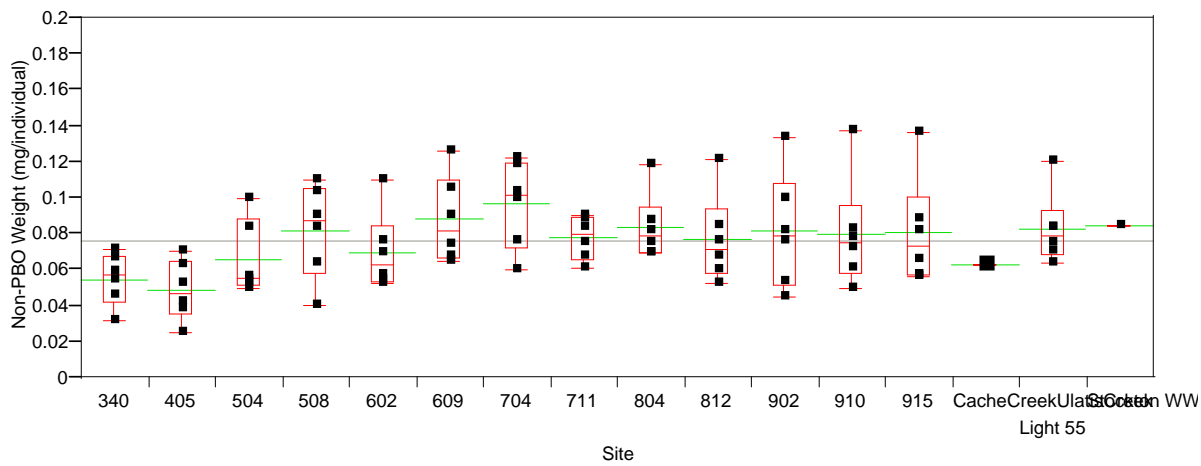
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Effects on Non-PBO Weight (measured as % of Non-PBO Control Weight)

Term	Scaled Estimate	Plot Estimate	Std Error	t Ratio	Prob> t
Intercept	128.7		6.6	19.40	<.0001
Log EC	5.0		13.2	0.38	0.7049
(Log EC) ²	-28.7		17.6	-1.63	0.1072
Sample[340]	20.1		13.0	1.55	0.1264
Sample[405]	-7.4		19.1	-0.39	0.7002
Sample[504]	0.4		14.5	0.03	0.9786
Sample[508]	5.6		14.2	0.40	0.6939
Sample[602]	5.1		15.3	0.33	0.7415
Sample[609]	4.2		14.6	0.29	0.7732
Sample[704]	-13.7		13.0	-1.05	0.2951
Sample[711]	-17.6		15.1	-1.17	0.2466
Sample[804]	11.0		14.0	0.79	0.4340
Sample[812]	-4.2		12.7	-0.33	0.7400
Sample[902]	2.1		12.5	0.17	0.8618
Sample[910]	-5.2		12.5	-0.42	0.6784
Sample[915]	0.5		12.5	0.04	0.9682
Sample[Light 55]	-0.9		12.7	-0.07	0.9433

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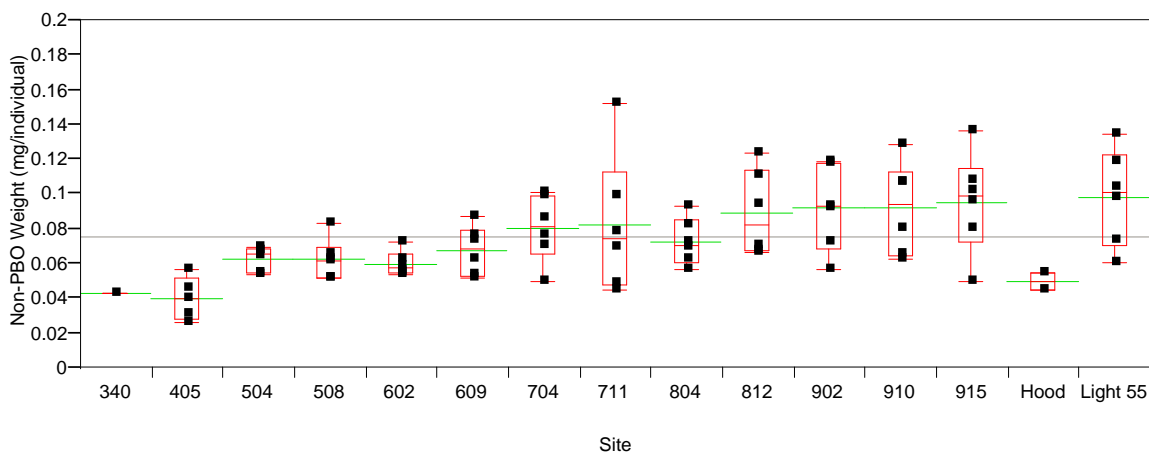


Effects on Non-PBO Weight (measured as % of Non-PBO Control Weight)

Term	Scaled Estimate	Plot Estimate	Std Error	t Ratio	Prob> t
Intercept	160.7		43.8	3.66	0.0011
Log EC	132.0		86.1	1.53	0.1373
(Log EC) ²	10.3		83.1	0.12	0.9020
Sample[340]	-158.9		82.6	-1.92	0.0654
Sample[405]	-202.3		88.5	-2.29	0.0307
Sample[504]	-71.8		72.0	-1.00	0.3276
Sample[508]	-14.3		69.0	-0.21	0.8365
Sample[602]	-114.5		74.3	-1.54	0.1356
Sample[609]	-21.8		68.1	-0.32	0.7518
Sample[704]	21.0		49.3	0.43	0.6735
Sample[711]	128.8		98.9	1.30	0.2041
Sample[804]	17.4		52.8	0.33	0.7444
Sample[812]	85.8		76.1	1.13	0.2704
Sample[902]	56.3		65.2	0.86	0.3960
Sample[910]	65.2		60.4	1.08	0.2901
Sample[915]	82.5		66.8	1.24	0.2276
Sample[Light 55]	126.5		88.3	1.43	0.1638

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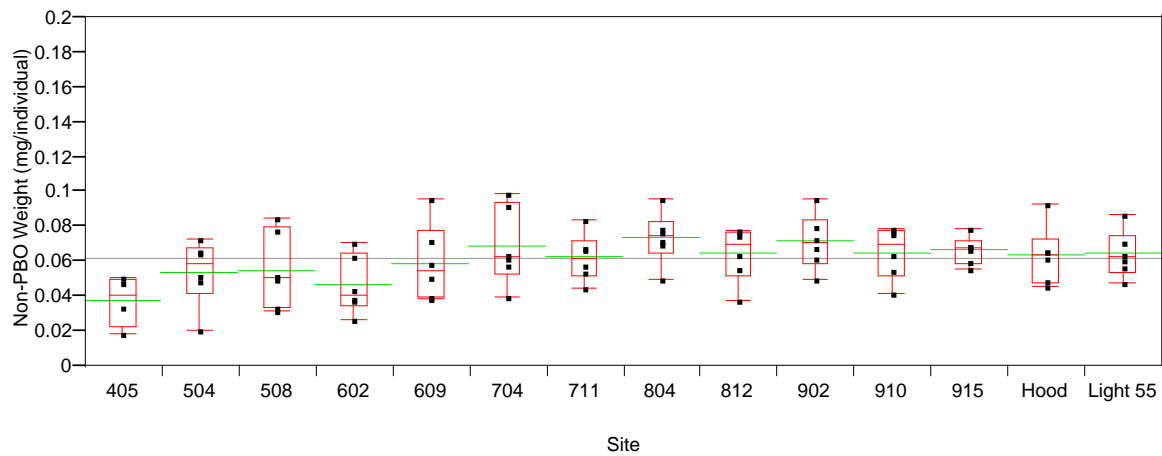
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Effects on Non-PBO Weight (measured as % of Non-PBO Control Weight)

Term	Scaled Estimate	Plot Estimate	Std Error	t Ratio	Prob> t
Intercept	105.8		32.5	3.26	0.0019
Log EC	79.9		55.1	1.45	0.1528
(Log EC) ²	139.0		67.9	2.05	0.0455
Sample[405]	-239.0		88.9	-2.69	0.0094
Sample[504]	-92.6		51.9	-1.78	0.0799
Sample[508]	-35.9		35.9	-1.00	0.3217
Sample[602]	-152.1		67.8	-2.25	0.0287
Sample[609]	-71.7		49.1	-1.46	0.1495
Sample[704]	74.1		36.1	2.05	0.0450
Sample[711]	25.9		51.6	0.50	0.6172
Sample[804]	45.2		34.3	1.32	0.1933
Sample[812]	82.0		44.7	1.84	0.0715
Sample[902]	93.6		43.6	2.15	0.0361
Sample[910]	87.0		45.0	1.94	0.0579
Sample[915]	95.9		43.9	2.18	0.0332
Sample[Light 55]	87.6		46.9	1.87	0.0667

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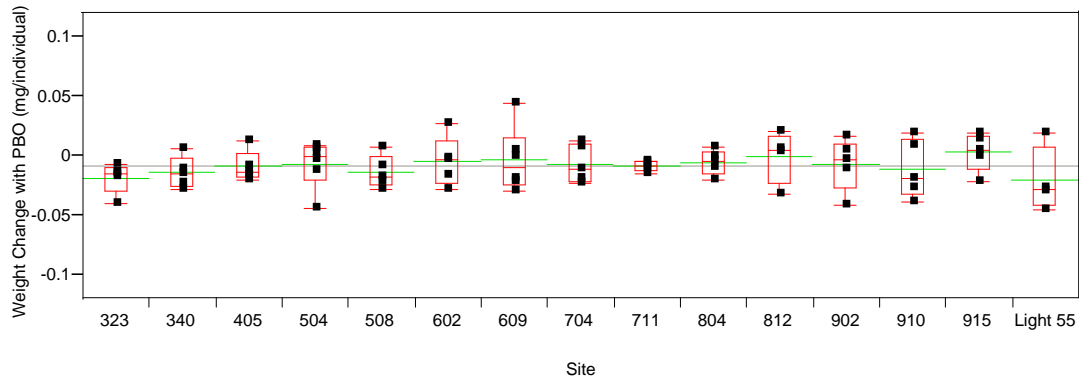
Effects on Non-PBO Weight (measured as % of Non-PBO Control Weight)

Term	Scaled Estimate	Plot Estimate	Std Error	t Ratio	Prob> t
Intercept	125.5		19.9	6.29	<.0001
Log EC	-84.8		49.3	-1.72	0.0904
(Log EC) ²	17.1		44.5	0.38	0.7024
Sample[405]	26.9		47.6	0.57	0.5733
Sample[504]	48.2		38.5	1.25	0.2151
Sample[508]	37.0		33.2	1.12	0.2682
Sample[602]	32.2		40.9	0.79	0.4332
Sample[609]	52.6		36.6	1.44	0.1549
Sample[704]	29.0		26.1	1.11	0.2694
Sample[711]	-71.6		48.4	-1.48	0.1436
Sample[804]	50.3		25.0	2.02	0.0478
Sample[812]	-16.3		23.0	-0.71	0.4807
Sample[902]	-2.3		23.4	-0.10	0.9231
Sample[910]	-29.2		27.7	-1.06	0.2945
Sample[915]	-16.8		24.6	-0.68	0.4963
Sample[Hood]	-75.8		51.3	-1.48	0.1441
Sample[Light 55]	-64.4		45.6	-1.41	0.1627

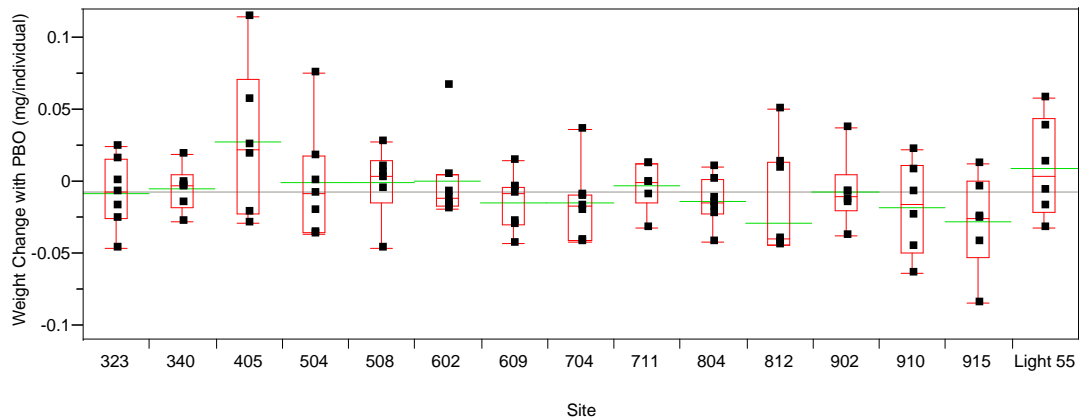
Appendix B. Section 2. *H. azteca* weight changes with PBO addition.

Weight changes induced by PBO addition in *H. azteca* 10 day water column toxicity tests did not differ significantly between sites in any season (one-way ANOVAs, $P > 0.05$). Nonetheless, box plots show some apparent variation between sites in the effects of PBO addition.

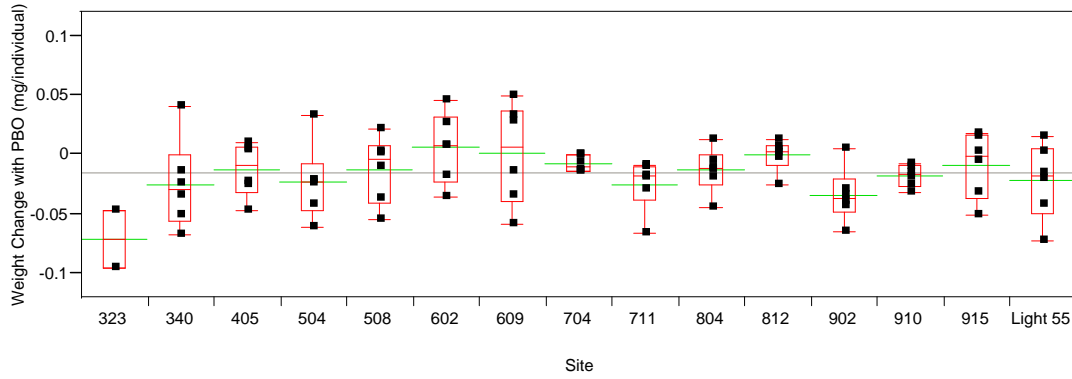
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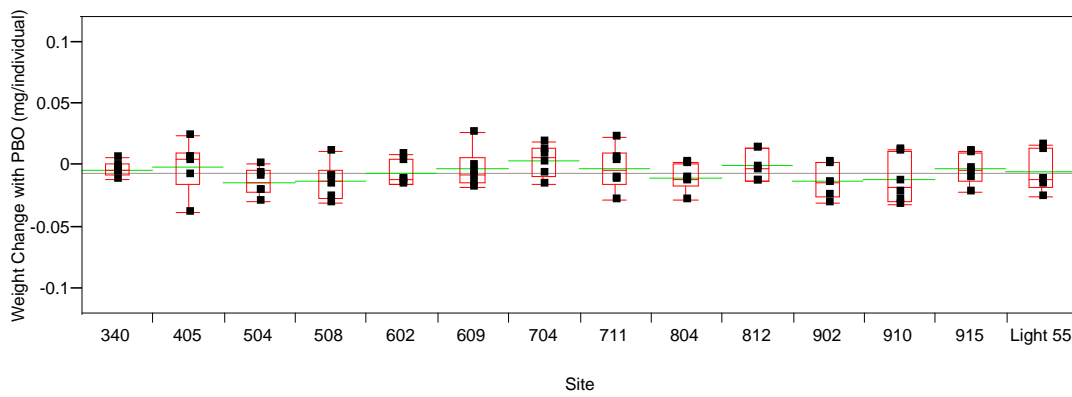
Spring 2006



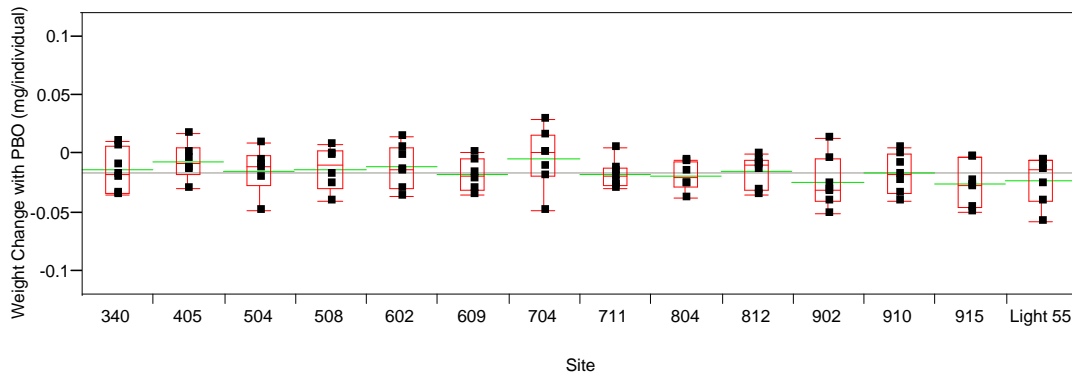
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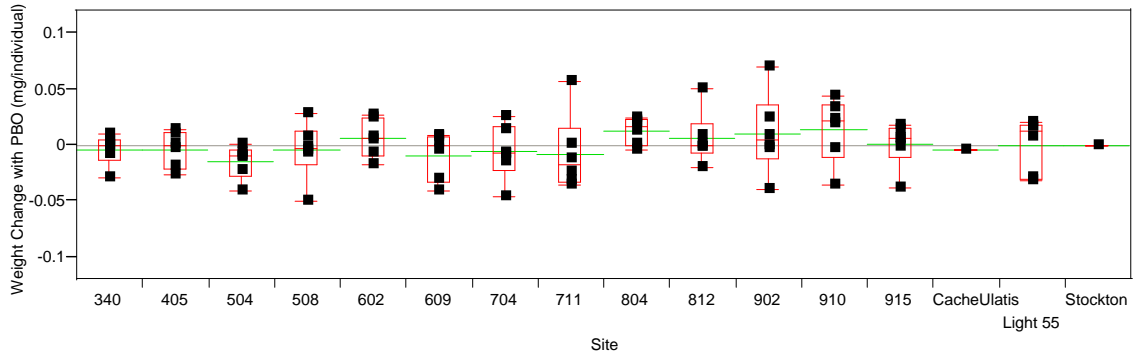


Winter 2007

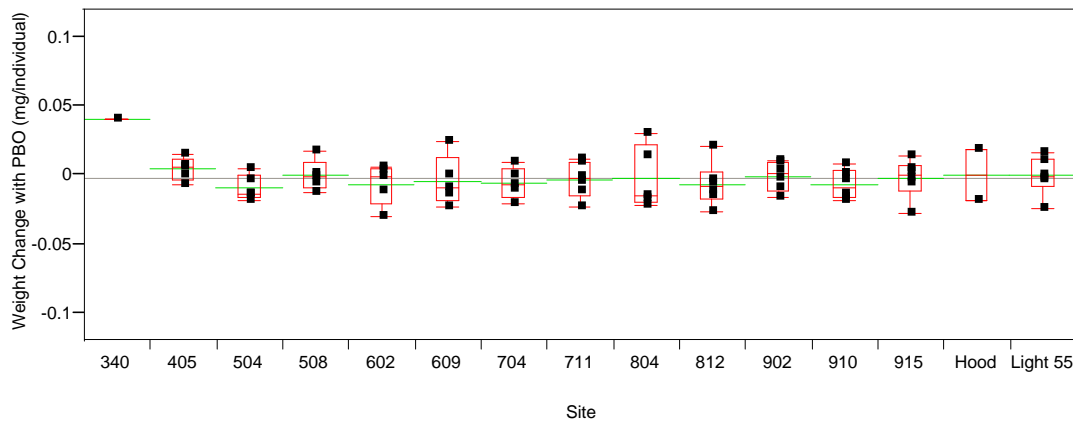


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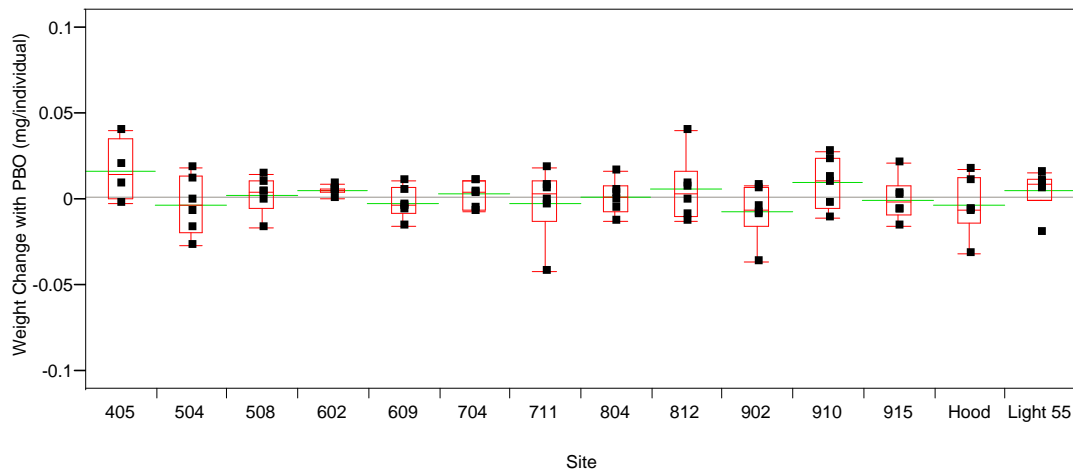
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Appendix C

Morone saxatilis

(Striped Bass)

Table C1-1. Results of 7-day *M. saxatilis* toxicity test initiated on 7/30/05 conducted on samples collected by UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 7/27/05 and 7/28/05.

Treatment	Survival (%) ¹		Length (cm) ¹		Weight (g) ¹	
	mean	se	mean	se	mean	se
Control	100 ^P	0.0	5.2	0.3	1.4	0.1
915	100	0.0	5.3	0.1	1.5	0.1
711	96	4.0	5.3	0.1	1.4	0.1
609	100	0.0	5.4	0.1	1.7	0.1
340	100	0.0	5.2	0.1	1.5	0.1

P. The laboratory control met the criteria for test acceptability.

1. Highlighted areas indicate a significant reduction in survival, length, or weight when compared to the laboratory control. All endpoints were analyzed according to EPA standard methods (1-tailed test, $P < 0.05$).

Table C1-2. Summary of water chemistry measurements taken during a 7-day *M. saxatilis* toxicity test initiated on 7/30/05 examining samples collected by UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 7/27/05 and 7/28/05.

Treatment	Lab Temp (°C)	Lab pH	Lab EC (µmhos/cm)	Lab DO (mg/L)	Hardness (mg/L CaCO ₃)	Alkalinity (mg/L CaCO ₃)	Ammonia Nitrogen (mg/L)
Control	21	7.98	414	10.2	208	222	NAP
915	19	8.03	157	8.8	96	70	0.00
711	20	7.89	132	8.8	92	78	0.07
609	20	7.63	2740	8.4	376	92	0.10
340	19	7.58	7640	8.8	188	144	0.01

Table C2-1. Effects of Cu+ on striped bass (*M. saxatilis*) during a 7-day exposure conducted using dilute well water spiked with copper chloride initiated on 8/11/05.

Total Cu ⁺ (2)	Dissolved Cu ⁺ (2)	Survival (%) ¹		Length (cm) ¹		Weight (g) ¹	
		mean	sd	mean	sd	mean	sd
0 ppb	0 ppb	100 ^P	0.0	5.48	0.34	1.73	0.40
42 ppb	40 ppb	100	0.0	5.41	0.29	1.58	0.30
160 ppb	160 ppb	92	17.9	5.42	0.41	1.66	0.49
470 ppb	440 ppb	0	0.0	-	-	-	-
900 ppb	810 ppb	0	0.0	-	-	-	-

Endpoint	Total Cu ⁺ (ug/L)			Dissolved Cu ⁺ (ug/L)		
	LC50	NOEC	LOEC	LC50	NOEC	LOEC
96 Hours	441	160	470	414	160	440
7 Days	262	160	470	254	160	440

P. The laboratory control met the criteria for test acceptability.

1. Highlighted areas indicate a significant reduction in survival, length, or weight when compared to the laboratory control. All endpoints were analyzed according to EPA standard methods. (1-tailed test, $P < 0.05$)

2. Copper concentrations were measured analytically.

Table C3-1. Summary of a Striped Bass (*M. saxatilis*) water column toxicity test initiated on 6/20/06 conducted on control water with test animals loaded using two alternative techniques.

Treatment	Day 1 Survival (%) ¹	Day 2 Survival (%) ¹	Day 4 Survival (%) ¹	Day 6 Survival (%) ¹	Day 7 Survival (%) ¹
Short cut pipette with elastic bulb	70	38	5	0	0
Long pipette with pipette pump	70	53	5	5	5
MSD	27	45	19	17	17

1. No significant differences in survival were found at any timepoint.

Table C3-2. Summary of water chemistry measurements during a Striped Bass (*M. saxatilis*) test initiated on 6/20/06 conducted on control water using two alternative loading techniques.

Treatment	Temp (°C)	pH	EC (µS/cm)	DO (mg/L)	Max Unionized Ammonia (mg/L)
Short cut pipette with elastic bulb	21	8.28	912	9.2	0.034
Long pipette with pipette pump	21	8.28	912	9.2	0.034

Table C4-1. Summary of a Striped Bass (*M. saxatilis*) water column toxicity test initiated on 7/14/06 conducted on samples collected by UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 7/11/06 - 7/12/06.

Treatment	Day 1 Survival (%) ¹	Day 2		Day 3			Day 4					
		Survival (%) ²		Survival (%) ²			Survival (%) ²					
		A	B	A	B	C	A	B	C			
Well Water	89	66	A	B	61	A	B	C	45	A	B	C
Low EC Control @ 100 μ S/cm	81	61		B	56	A	B	C	33	A	B	C
High EC Control @ 18,500 μ S/cm	97	97	A		83	A	B		75	A	B	
POD 508	94	88	A	B	83	A	B	C	74	A	B	
POD 915	79	68	A	B	44		B	C	28		B	C
POD 910	93	84	A	B	64	A	B	C	34	A	B	C
POD 711	90	69	A	B	43			C	24			C
POD 340 ³	100	97	A		95	A			82	A		
POD 609	100	91	A	B	82	A	B	C	64	A	B	C
MSD	41	47			53				65			

1. No significant differences in survival were found at this timepoint.
2. Treatments not sharing at least one significance group are significantly different from one another using Tukey's multiple comparison procedure.
3. This High EC sample should be compared to the High EC control.

Table C4-2. Summary of water chemistry measurements during a Striped Bass (*M. saxatilis*) test initiated on 7/14/06 conducted on samples collected by UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 7/11/06 - 7/12/06.

Treatment	Laboratory Chemistry				Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Initial Un-ionized Ammonia (mg/L)	Maximum Un-ionized Ammonia (mg/L)
	Temp (°C)	pH	EC (μ S/cm)	DO (mg/L)				
Well Water	22	8.10	675	9.4	NA	NA	NA	0.009
Low EC Control @ 100 μ S/cm	21	8.24	128	8.9	NA	NA	NA	0.015
High EC Control @ 18,500 μ S/cm	21	7.77	16490	9.0	NA	NA	NA	0.003
POD 508	20	7.72	877	9.5	122	52	0.000	0.006
POD 915	20	7.70	146	8.9	42	40	0.001	0.006
POD 910	20	7.32	269	7.5	72	52	0.003	0.004
POD 711	20	7.75	156	9.8	52	56	0.003	0.008
POD 340	20	7.44	15750	9.2	1960	160	0.001	0.003
POD 609	19	7.50	650	8.7	102	70	0.001	0.003

Table C5-1. Summary of a 7-day Striped Bass (*M. saxatilis*) water column toxicity test initiated on 8/25/06 examining samples collected by UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 8/22/06 - 8/23/06.

Treatment	Survival (%) ¹		Weight (g) ¹		Length (cm) ¹	
	Mean	SE	Mean	SE	Mean	SE
Well Water	100	0.0	4.09	0.38	7.0	0.18
High EC Control @ 18 mS/cm	100	0.0	3.14	0.36	6.4	0.21
Suisun Bay at Middle Point (508)	100	0.0	3.64	0.44	6.6	0.19
Montezuma Slough at Nurse Slough (609)	100	0.0	3.62	0.20	6.6	0.12
Old River at Holland Cut (915)	100	0.0	4.39	0.57	7.0	0.31
San Joaquin River at Hog and Turner Cut (910)	100	0.0	3.20	0.18	6.4	0.10
Sacramento River at tip of Grand Island (711)	100	0.0	4.08	0.12	6.8	0.06
Napa River at Vallejo Seawall (340) ²	100	0.0	4.35	0.09	7.0	0.09
Field Duplicate: Montezuma Slough at Nurse Slough (609)	100	0.0	4.92	0.36	7.4	0.18

Survival PMSD: NA³

Weight PMSD: 53.0

Length PMSD: 16.2

1. No significant differences in survival, weight or length were found compared to the appropriate control.
2. This high conductivity sample was compared to the High EC control.
3. Not possible to calculate MSD due to lack of variability in data.

Table C5-2. Summary of water chemistry measurements during a 7-day striped bass (*M. saxatilis*) test initiated on 8/25/06 examining samples collected by UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 8/22/06 - 8/23/06.

Treatment	Laboratory Chemistry				Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Initial Unionized Ammonia (mg/L)	Maximum Unionized Ammonia (mg/L)
	EC (μS/cm)	Temp (°C)	pH	DO (mg/L)				
Well Water	729	20.4	7.88	9.2	NA	NA	NA	0.653
High EC Control @ 18,000 uS/cm	18800	20.4	7.76	8.9	NA	NA	NA	0.236
Suisun Bay at Middle Point (508)	3007	20.0	7.65	9.2	396	68	0.002	0.160
Montezuma Slough at Nurse Slough (609)	4887	20.3	7.53	8.9	720	85	0.002	0.153
Old River at Holland Cut (915)	222	19.7	7.73	9.1	68	58	0.001	0.435
San Joaquin River at Hog and Turner Cut (910)	343	19.7	7.17	7.9	100	74	0.003	0.306
Sacramento River at tip of Grand Island (711)	133	20.0	7.59	9.2	56	64	0.005	0.227
Napa River at Vallejo Seawall (340)	16070	20.1	7.46	8.7	3720	260	0.000	0.175
Field Duplicate: Montezuma Slough at Nurse Slough (609)	5010	20.0	7.54	8.9	670	85	0.002	0.128

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Table C6-1. Effects of esfenvalerate on striped bass (*M. saxatilis*) during a 24-hour exposure initiated on 8/22/06.

Treatment ²	4 hours							
	Survival (%)		Normal Swimming (%) ¹					
	Mean	SE	Mean	SE				
Control	1.00	0.00	1.00	0.00				
Solvent Control	1.00	0.00	1.00	0.00				
0.64 ug/L Esfenvalerate	1.00	0.00	1.00	0.00				
2.2 ug/L Esfenvalerate	1.00	0.00	1.00	0.00				
4.4 ug/L Esfenvalerate	1.00	0.00	0.64	0.07				
6.4 ug/L Esfenvalerate	1.00	0.00	0.24	0.04				

Treatment ²	24 hours							
	Survival (%) ¹		Normal Swimming (%) ¹		Length (cm)		Weight (g)	
	Mean	SE	Mean	SE	Mean	SE	Mean	SE
Control	1.00	0.00	1.00	0.00	6.58	0.42	3.74	0.74
Solvent Control	1.00	0.00	0.96	0.04	6.44	0.39	3.46	0.61
0.64 ug/L Esfenvalerate	1.00	0.00	1.00	0.00	6.65	0.36	3.91	0.60
2.2 ug/L Esfenvalerate	0.40	0.19	0.28	0.19	6.62	0.55	4.02	1.06
4.4 ug/L Esfenvalerate	0.00	0.00	0.00	0.00	-	-	-	-
6.4 ug/L Esfenvalerate	0.00	0.00	0.00	0.00	-	-	-	-

1: Highlighted areas indicate a significant reduction in survival or normal swimming behavior when compared to the laboratory control. All endpoints were analyzed according to EPA standard methods (1-tailed test, P < 0.05)

2. Esfenvalerate concentrations were measured analytically.

Time	Survival			Swimming Behavior		
	LC50 ³	NOEC ³	LOEC ³	EC25 ³	NOEC ³	LOEC ³
4 hours	NA	6.4	> 6.5	3.88	2.2	4.4
24 hours	2.17	0.64	2.2	1.07	0.64	2.2

3: all concentrations given in ug/L Esfenvalerate

Appendix D

Hypomesus transpacificus

(Delta Smelt)

Methods Tests

Table D1-1. Summary of a 7-day *H. transpacificus* test initiated on 8/11/05 examining survival in three different holding conditions.

Treatment	Rep	# Surviving Fish by Test Day						
		1	2	3	4	5	6	7
5 fish in 5 Liters	1	4	4	4	4	4	3	3
	2	4	3	3	3	3	3	3
Ammonia-N (mg/L)		0.19	0.94	1.14	0.39	1.24	1.87	0.79
10 fish in 5 Liters	1	7	7	7	7	7	7	7
	2	10	9	9	9	9	9	9
Ammonia-N (mg/L)		0.34	1.27	1.42	0.42	1.78	2.75	1.30
10 fish in 8 Liters	1	10	10	10	10	10	10	10
	2	10	9	9	9	9	9	9
Ammonia-N (mg/L)		0.25	0.91	1.05	0.38	1.21	2.15	0.72
% Water Renewed		50	50	80	50	50	80	

Table D1-2. Summary of water chemistry measurements taken on termination of *H. transpacificus* holding conditions test initiated on 8/11/05.

Treatment	Temp (°C)	pH	EC (uS/cm)	DO (mg/L)	Ammonia Nitrogen (mg/L)
5 fish in 5 liters	21	8.29	456	8.4	0.79
10 fish in 5 liters	21	8.42	458	8.9	1.30
10 fish in 5 liters	20	8.34	451	8.9	0.72

Table D2-1. Summary of a *H. transpacificus* water column toxicity test initiated on 4/18/06 examining the response of 15 day old fish to control waters with algae added.

Water Bath	Treatment	Day 2 Survival ¹		Day 4 Survival ¹		Day 6 Survival ¹		Day 7 Survival ¹		Day 7 Biomass ¹	
		Survival (%)	Significance Group	Survival (%)	Significance Group	Survival (%)	Significance Group	Survival (%)	Significance Group	Biomass (mg/individual)	Significance Group
1	Hatchery Water	100	A	98	A	91	A	85	A	0.295	A
	DIEPAMH No Algae	83	B	58	A B C	NA	- - -	48	B	0.113	B
	DIEPAMH + 1x Algae	100	A	75	A B	45	B	13	B C	0.016	B
	DIEPAMH + 2x Algae	100	A	40	B C	0	C	0	C	0.000	B
	DIEPAMH + 3x Algae	95	A B	13	C	3	C	0	C	0.000	B
MSD	21		61		55		51		0.159		
2	Hatchery Water	100	A	100	A	100	A	92	A	0.299	A
	DIEPAMH No Algae	81	A	52	B	NA	- -	36	B	0.081	B
	DIEPAMH + 1x Algae	100	A	36	B	11	B	5	B	0.009	B
	DIEPAMH + 2x Algae	100	A	36	B	3	B	0	B	0.000	B
	DIEPAMH + 3x Algae	100	A	15	B	5	B	0	B	0.000	B
MSD	33		60		20		54		0.122		

1. Treatments not sharing at least one significance group are significantly different from one another using Tukey's multiple comparison procedure.

Table D2-2. Summary of water chemistry measurements during a *H. transpacificus* test initiated on 4/18/06 performed on control waters with algae added to increase turbidity.

Light	Treatment	Temp (°C)	pH	EC (uS/cm)	DO (mg/L)	Initial Unionized Ammonia (mg/L)	Maximum Unionized Ammonia (mg/L)
Fluorescent	Hatchery Water	19	7.32	1654	9.5	0.009	0.013
	DIEPAMH No Algae	19	8.01	288	9.5	0.001	0.016
	DIEPAMH + 1x Algae	18	8.00	289	8.5	0.003	0.029
	DIEPAMH + 2x Algae	20	7.75	291	8.6	0.000	0.031
	DIEPAMH + 3x Algae	20	7.87	306	8.4	0.000	0.060
Ambient	Hatchery Water	19	6.55	1689	8.4	0.003	0.015
	DIEPAMH No Algae	19	8.01	288	9.5	0.001	0.012
	DIEPAMH + 1x Algae	18	8.00	289	8.5	0.004	0.018
	DIEPAMH + 2x Algae	19	8.02	291	9.3	0.004	0.035
	DIEPAMH + 3x Algae	20	7.87	306	8.4	0.003	0.042

Ambient Monitoring Toxicity Tests – 2005

Table D3-1. Summary of 7-day *H. transpacificus* toxicity test conducted on samples collected by the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 8/30/05.¹

Treatment	Survival (%) ²		Length (cm) ²		Weight (g) ²	
	mean	se	mean	se	mean	se
Laboratory Control (Dilute Well Water)	95 ^P	2.9	3.79	0.06	0.29	0.01
711	98	2.5	3.87	0.09	0.29	0.02
915	92	4.9	3.94	0.08	0.34	0.03
609	100	0.0	3.91	0.02	0.32	0.00
340	85	2.9	3.97	0.06	0.32	0.02

P. The laboratory control met the criteria for test acceptability.

1. This test was set up on 9/01/05.

2. Highlighted areas indicate a significant reduction in survival, length, or weight when compared to the laboratory control. All endpoints were analyzed according to EPA standard methods. (1-tailed test, $P < 0.05$)

Table D3-2. Summary of water chemistry measurements taken during a 7-day *H. transpacificus* toxicity test initiated on 9/01/05 examining samples collected by the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 8/30/05.

Treatment	Temp (°C)	pH	EC (uS/cm)	DO (mg/L)	Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Ammonia Nitrogen (mg/L)
Lab Control (Dilute Well Water)	23	8.16	470	8.7	212	200	NAP
711	21	7.91	169	9.1	68	78	0.13
915	21	8.04	360	9.1	740	70	0.07
609	22	7.58	6460	8.2	2236	100	0.08
340	22	7.55	19050	8.4	80	138	0.03

Ambient Monitoring Toxicity Tests – 2006

Table D4-1. Summary of a 7-day *H. transpacificus* water column toxicity test initiated on 4/05/06 conducted on samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) on 4/03/06 - 4/04/06. (9 day old smelt)

Treatment	Day 2 Survival (%) ¹	Day 4 Survival ²		Day 6 Survival ²		Day 7 Survival (%) ¹
		Survival (%)	Significance Group	Survival (%)	Significance Group	
Lab Control (DIEPAMH)	92	23	B	0	B	0
340	100	75	A	45	A	8
508	100	75	A	5	B	0
609	93	85	A	12	B	0
711	100	93	A	15	B	0
910	87	82	A	5	B	0
915	98	87	A	16	B	0
MSD	19	46		22		11

1. No significant differences in survival were found at this timepoint
2. Treatments not sharing at least one significance group are significantly different from one another using Tukey's multiple comparison procedure.

Table D4-2. Summary of water chemistry measurements during a *H. transpacificus* test performed on samples collected by the the UC Davis Aquatic Toxicology and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 4/05/06.

Treatment	Temp (°C)	pH	EC (uS/cm)	DO (mg/L)	Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Initial Unionized Ammonia (mg/L)	Maximum Unionized Ammonia (mg/L)
Lab Control (DIEPAMH)	18	7.99	290	9.6	84	64	-	0.019
340	18	7.57	815	9.8	140	76	0.004	0.008
508	17	7.63	220	10.6	68	56	0.002	0.014
609	18	7.43	574	10.5	128	78	0.002	0.018
711	18	7.48	141	10.8	52	48	0.002	0.023
910	18	7.67	208	10.7	60	54	0.007	0.017
915	18	7.61	182	10.7	60	48	0.002	0.009

Table D5-1. Summary of a 7-day *H. transpacificus* water column toxicity test conducted on samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) on 5/01/06 - 5/02/06.¹ (30 day old Smelt)

Treatment	Day 2 Survival (%) ²	Day 4 Survival (%) ²	Day 6 Survival ³			Day 7 Survival ³				
			Survival (%)	Significance Group		Survival (%)	Significance Group			
Hatchery Water from Fish Culture ⁴	98	95	79	A		79	A			
Hatchery Water from Tap ⁴	100	79	54	A	B	C	43	B	C	D
Diluted Hatchery Water	100	92	63	A	B	C	53	A	B	C
711	98	82	58	A	B	C	40	B C D		
910	95	57	51	A	B	C	30	C D		
915	98	76	37	B C			27	C D		
340	95	71	61	A	B	C	55	A	B	C
508	95	78	28	C			18	D		
609	100	95	73	A	B		64	A	B	
MSD	16	51	48				45			

1. This test was set up on 5/03/06.
2. No significant differences in survival were found at this timepoint
3. Treatments not sharing at least one significance group are significantly different from one another using Tukey's multiple comparison procedure.
4. Hatchery water from the fish culture was Hatchery Tap Water with Nanno 3600 and salt water added.

Table D5-2. Summary of water chemistry measurements during a 7-day *H. transpacificus* test initiated on 5/03/06 examining samples collected by the the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 5/01/06 - 5/02/06.

Treatment	Temp (°C)	pH	EC (uS/cm)	DO (mg/L)	Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Initial Unionized Ammonia (mg/L)	Maximum Unionized Ammonia (mg/L)
Hatchery Water	17.1	7.19	1117	8.9	-	-	0.001	0.003
Filtered Hatchery Water	17.1	7.84	135	9.7	-	-	0.000	0.008
Diluted Hatchery Water	17.4	7.46	154	9.6	-	-	0.000	0.005
711	19.3	7.84	104	9.5	52	46	0.003	0.015
910	17.6	7.66	137	8.9	44	40	0.002	0.011
915	17.0	7.31	152	9.0	44	42	0.001	0.009
340	17.3	7.64	461	9.1	124	68	0.002	0.014
508	17.3	7.80	141	9.2	56	58	0.002	0.018
609	17.6	7.80	506	9.1	112	70	0.004	0.014

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Table D6-1. Summary of a *H. transpacificus* water column toxicity test conducted on samples collected by the UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) on 5/15/06 - 5/16/06.¹ (40 day old Smelt)

Treatment	Day 1 Survival ²			Day 2 Survival ²		
	Survival (%)	Significance Group		Survival (%)	Significance Group	
Hatchery Water	35	A	B	5	C	
711	44	A	B	33	B	C
910	50	A	B	35	A	B C
915	76	A	B	24	B	C
340	83	A		74	A	
508	30		B	5		C
609	70	A	B	50	A	B
Low Salinity Hatchery Water + Algae	NA	-	-	10	B	C
MSD	69			52		

1. This test was set up on 5/17/06.
2. Treatments not sharing at least one significance group are significantly different from one another using Tukey's multiple comparison procedure.

Table D6-2. Summary of water chemistry measurements during a *H. transpacificus* test performed on samples collected by the UC Davis Aquatic Toxicology Laboratory (UCD ATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 5/15/06 - 5/16/06.

Treatment	Temp (°C)	pH	EC (uS/cm)	DO (mg/L)	Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Initial Unionized Ammonia (mg/L)	Maximum Unionized Ammonia (mg/L)
Hatchery Water	18.2	7.50	703	9.3	-	-	-	0.009
711	18.0	7.74	102	10.3	44	46	0.003	0.009
910	18.2	7.85	134	9.8	44	37	0.007	0.009
915	19.0	7.48	130	9.6	36	35	0.002	0.008
340	18.6	7.61	2347	9.7	280	72	0.003	0.007
508	18.6	7.96	127	9.6	52	46	0.007	0.009
609	18.2	8.10	314	9.3	72	58	0.010	0.011
Low Salinity Hatchery Water + Algae	17.9	7.81	112	8.9	-	-	-	0.005

Table D7-1. Summary of a *H. transpacificus* water column toxicity test conducted on samples collected by the UC Davis Aquatic Toxicology Laboratory (UCD ATL) and the California Department of Fish and Game (CDFG) on 5/30/06 - 5/31/06.¹ (60 day old Smelt)

Treatment	Day 2 Survival ²		Day 4 Survival ²	
	Survival (%)	Significance Group	Survival (%)	Significance Group
Hatchery Water	60	A B	55	A B
Low Salinity Hatchery Water + Algae	37	B	30	A B
711	38	B	20	B
910	45	A B	33	A B
915	43	A B	31	B
340	88	A	86	A
508	68	A B	58	A B
609	68	A B	68	A B
MSD	62		69	

1. This test was set up on 6/01/06.
2. Treatments not sharing at least one significance group are significantly different from one another using Tukey's multiple comparison procedure.

Table D7-2. Summary of water chemistry measurements during a *H. transpacificus* test performed on samples collected by the UC Davis Aquatic Toxicology Laboratory (UCD ATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 5/30/06 - 5/31/06.

Treatment	Temp (°C)	pH	EC (uS/cm)	DO (mg/L)	Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Initial Unionized Ammonia (mg/L)	Maximum Unionized Ammonia (mg/L)
Hatchery Water	16	7.99	491	10.1	-	-	0.001	0.005
Low Salinity Hatchery Water + Algae	17	8.20	124	9.8	-	-	0.006	0.021
711	16	7.94	124	10.5	50	52	0.008	0.026
910	15	7.80	131	10.8	40	35	0.006	0.015
915	15	7.68	1355	10.8	32	34	0.003	0.014
340	16	7.66	3412	10.0	382	61	0.003	0.013
508	13	7.55	614	10.3	48	46	0.001	0.020
609	14	7.69	624	9.9	92	60	0.004	0.014

Table D8-1. Summary of a *H. transpacificus* water column toxicity test conducted on samples collected by the UC Davis Aquatic Toxicology Laboratory (UCD ATL) and the California Department of Fish and Game (CDFG) on 6/13/06 - 6/14/06.¹ (90 day old Smelt)

Treatment	Day 1 Survival (%) ²	Day 2 Survival (%) ²	Day 4 Survival (%) ²	Day 6 Survival (%) ²	Day 7 Survival (%) ²
Hatchery Water (1500 uS/cm)	96	88	75	58	58
711 salted to 1500 uS/cm	83	67	67	63	59
Low Conductivity Control	83	71	62	62	62
711	92	88	69	66	66
910	84	64	50	35	35
915	85	77	52	41	41
340	96	96	77	78	81
508	77	77	52	43	44
609	79	71	50	46	46
Hatchery Water (1500 uS/cm) in 2 nd water bath	83	80	66	60	62
DIEPAMH salted to 1500 uS/cm in 2 nd water bath	91	91	82	77	77
MSD	51	71	80	77	76

1. This test was set up on 6/15/06.
2. No significant differences in survival were found at any timepoint

Table D8-2. Summary of water chemistry measurements during a *H. transpacificus* test performed on samples collected by the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 6/13/06 -6/14/06.

Treatment	Temp (°C)	pH	EC (uS/cm)	DO (mg/L)	Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	Initial Unionized Ammonia (mg/L)	Maximum Unionized Ammonia (mg/L)
Hatchery Water (1500 uS/cm)	19	7.90	1535	9.2	40	32	0.004	0.011
711 salted to 1500 uS/cm	19	7.93	1474	9.6	172	57	0.006	0.012
Low Conductivity Control	18	7.83	241	8.9	12	10	0.004	0.011
711	18	8.05	120	9.3	48	48	0.009	0.012
910	19	7.88	124	8.9	42	32	0.005	0.011
915	18	7.85	179	9.1	52	36	0.003	0.012
340	21	7.76	6400	8.9	876	70	0.005	0.011
508	18	8.09	137	9.4	44	48	0.010	0.013
609	18	8.11	195	9.4	60	52	0.005	0.010
Hatchery Water (1500 uS/cm) in 2 nd water bath	19	7.90	1535	9.2	40	32	0.004	0.012
DIEPAMH salted to 1500 uS/cm in 2 nd water bath	18	8.05	1493	9.5	190	60	0.003	0.006

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Ambient Monitoring Toxicity Tests – 2007

Table D9-1. Summary of a 7-day *H. transpacificus* water column toxicity test initiated on 5/12/07 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 5/8/07 and 5/10/07.¹

Treatment ³	Survival (%) ²													Weight ² (mg/surviving individual)		Biomass ² (mg/original individual)						
	Day 1	Day 2	Day 3	Day 4	Day 5	5A	5B	Day 6	6A	6B	6C	Day 7	7A	7B	7C	Mean	SE	Mean	SE	A	B	C
Low EC Control @ 180 uS/cm	98	87	82	71	64			33			C	21			C	0.948	0.072	0.195	0.084			C
711	100	82	71	59	48		B	30			C	20			C	0.889	0.173	0.173	0.067			C
Hood	100	87	81	74	69			50		B	C	45		B	C	0.824	0.069	0.384	0.101			
915	95	93	88	83	78			68				45		B	C	0.909	0.064	0.410	0.084			
Vernalis	100	90	80	70	62			54				45		B	C	0.821	0.066	0.362	0.062			
Hatchery Rearing Water @ 2500 uS/cm	95	93	88	86	84			60				39		B	C	0.767	0.047	0.299	0.035			B C
High EC Control @ 5000 uS/cm	100	97	97	97	89		A	89		A	B	73		A	B	0.813	0.062	0.597	0.074	A	B	
609	95	78	78	76	68			59				51				0.832	0.051	0.404	0.082			
508	100	95	85	85	70			54				43		B	C	0.795	0.063	0.352	0.092			B C
340	100	94	94	91	91		A	91		A		89		A		0.817	0.058	0.717	0.029	A		

1. Test was conducted in beakers.

Smelt were 36-days old at test initiation.

2. All samples and controls were compared to one another using ANOVA and Tukey's multiple comparison procedure.

3. The Low EC control showed lower Day 6 Survival, Day 7 Survival, and Biomass than the High EC Control.

Table D9-2. Water chemistry at field conditions of samples collected by the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 5/8/07 and 5/10/07.

Treatment	Field Chemistry				
	SC (uS/cm)	EC (uS/cm)	Temp (°C)	DO (mg/L)	pH
Sacramento R. at Grand Island (711)	177.7	157.8	19.4	8.60	8.05
Sacramento R. at Hood DWR Station	189.1	170.9	20.2	8.61	7.34
Old River at Holland Cut (915)	277.0	254.8	21.0	8.40	8.17
Sacramento R. at Vernalis DWR Station	474.0	436.1	21.0	9.85	8.06
Montezuma Slough at Nurse Slough (609)	4713.0	4043.8	17.9	8.64	7.88
Suisun Bay off Chipps Island (508)	5390.0	4678.5	18.4	9.76	8.02
Napa River at Riverside Blvd Terminus (340)	9640.0	9215	22.8	13.07	8.26

Table D9-3. Summary of water chemistry during a *H. transpacificus* (Delta Smelt) 7-day test initiated on 5/12/07 evaluating the toxicity of Sacramento River and Delta water samples collected by the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 5/8/07 and 5/10/07.

Treatment	Temp (°C)			EC (uS/cm)			DO (mg/L)			pH			Turbidity (NTU)			Un-ionized Ammonia (mg/L)			Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)
	Mean	SD	N	Mean	SD	N	Mean	SD	N	Mean	SD	N	Mean	SD	N	Mean	SD	N		
Low EC Control @ 160 uS/cm	16.4	0.4	8	157	4	4	8.7	0.9	8	7.70	0.24	8	-	-	0	0.009	0.001	3	-	-
Sacramento R. at Grand Island (711)	16.3	0.4	8	149	9	4	9.2	1.0	8	7.69	0.09	8	5.6	0.2	3	0.015	0.002	3	68	68
Sacramento R. at Hood DWR Station	16.5	0.4	8	156	5	4	9.2	0.9	8	7.61	0.13	8	4.2	-	1	0.021	0.002	3	72	66
Old River at Holland Cut (915)	16.6	0.5	8	236	8	4	9.1	1.0	8	7.70	0.15	8	3.9	0.0	3	0.011	0.001	3	80	66
Sacramento R. at Vernalis DWR Station	16.6	0.6	8	326	3	4	9.3	1.0	8	7.76	0.15	8	4.4	-	1	0.010	0.001	3	100	58
Hatchery Water @ 2500 uS/cm	16.7	0.6	8	2072	36	4	8.6	0.8	8	7.76	0.19	8	-	-	0	0.012	0.001	3	480	360
High EC Control @ 5000 uS/cm	16.7	0.8	8	4331	42	4	8.8	0.8	8	7.65	0.19	8	-	-	0	0.008	0.001	3	-	-
Montezuma Slough at Nurse Slough (609)	16.6	0.7	8	3834	33	4	9.2	0.9	8	7.63	0.09	8	40.2	0.8	3	0.010	0.002	3	540	88
Suisun Bay off Chipps Island (508)	16.6	0.7	8	4391	89	4	9.2	0.9	8	7.69	0.11	8	18.5	0.7	3	0.009	0.001	3	428	72
Napa River at Riverside Blvd Terminus (340)	16.6	0.6	8	7773	90	4	9.2	1.2	8	7.79	0.13	8	19.5	0.5	3	0.013	0.003	3	356	122

Table D10-1. Summary of a 7-day *H. transpacificus* water column toxicity test conducted in aquaria initiated on 5/24/07 examining samples collected by the UC Davis Aquatic Toxicology Laboratory (UCD ATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 5/23/07 and 5/24/07.

Treatment	Day 7 Survival (%) ¹					Weight (mg/surviving individual) ¹		Biomass (mg/original individual) ¹				
	mean	se	A	B	C	mean	se	mean	se	A	B	C
Low EC Control @ 180 uS/cm	31.8	6.6			C	0.742	0.039	0.233	0.047			C
Sacramento R. at tip of Grand Island (711)	36.5	4.0			C	0.633	0.061	0.226	0.021			C
Sacramento R. at Hood DWR Station	40.5	13.3		B	C	0.675	0.040	0.282	0.101			C
Old River at Holland Cut (915)	31.7	2.8			C	0.812	0.044	0.258	0.027			C
San Joaquin R. at Vernalis DWR Station	39.2	5.9		B	C	0.687	0.049	0.263	0.029			C
Hatchery Rearing Water @ 2188 uS/cm	53.0	7.1		B	C	0.895	0.150	0.462	0.066		B	C
High EC Control @ 5000 uS/cm	50.8	10.0		B	C	0.791	0.027	0.405	0.083		B	C
Montezuma Slough @ Nurse Slough (609) ²	90.0	0.9	A			0.840	0.039	0.756	0.031	A		
Sacramento R. across from Sherman Lake (508)	69.9	2.7	A	B		0.790	0.045	0.553	0.039	A	B	
Napa River at Riverside Blvd Terminus (340) ²	87.2	5.1	A			0.854	0.042	0.743	0.049	A		

1. No significant reduction in survival, weight or biomass were observed compared to the appropriate control. No significant differences in weight were observed between treatments. All pairwise comparisons were evaluated with Tukey's multiple comparison procedure.
2. Samples 609 and 340 showed significantly higher survival and biomass than the high EC control. Smelt were 30 days old at test initiation.

Table D10-2. Water chemistry at field conditions of samples collected by the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 5/23/07 and 5/24/07.

Sample	Field Chemistry					Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)
	SC (µS/cm)	EC (µS/cm)	Temp (°C)	DO (mg/L)	pH		
Sacramento R. at Grand Island (711)	202.3	186.9	21.2	9.0	7.92	56	62
Sacramento R. at Hood DWR Station	327.7	307.4	21.9	7.6	7.32	60	68
Old River at Holland Cut (915)	380.4	347.7	20.7	8.6	8.14	84	72
Sacramento R. at Vernalis DWR Station	451.9	403.1	19.6	11.0	8.39	100	64
Montezuma Slough at Nurse Slough (609)	5400.0	4762.8	19.1	8.1	7.80	300	80
Suisun Bay off Chipps Island (508)	3459.0	3037.0	18.9	9.2	8.05	380	90
Napa River at Riverside Blvd Terminus (340)	13490.0	12653.6	21.9	9.8	7.92	1520	160

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Table D10-3. Summary of water chemistry during a *H. transpacificus* (Delta Smelt) 7-day test initiated on 5/24/07 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 5/23/07 and 5/24/07.

Treatment	Temp (°C)			EC (µS/cm)			DO (mg/L)			pH			Turbidity (NTU)			Un-ionized Ammonia (mg/L)		
	Mean	SD	N	Mean	SD	N	Mean	SD	N	Mean	SD	N	Mean	SD	N	Mean	SD	N
Low EC Control @ 160 µS/cm	16.6	0.4	5	190	54	5	9.6	0.5	5	7.48	0.25	5	9.9	-	1	0.001	0.001	4
Sacramento R. at Grand Island (711)	16.3	0.4	5	194	55	5	9.7	0.2	5	7.82	0.07	5	5.7	-	1	0.012	0.003	5
Sacramento R. at Hood DWR Station	16.4	0.3	5	198	50	5	9.6	0.3	5	7.77	0.12	5	2.8	-	1	0.011	0.004	5
Old River at Holland Cut (915)	16.4	0.3	5	317	39	5	9.8	0.3	5	7.85	0.18	5	4.5	-	1	0.002	0.002	4
Sacramento R. at Vernalis DWR Station	16.4	0.5	5	379	55	5	9.7	0.2	5	7.92	0.08	5	5.9	-	1	0.002	0.002	5
Hatchery Water @ 2500 µS/cm	16.9	1.0	5	1411	647	5	9.6	0.2	5	7.84	0.13	5	19.7	-	1	0.003	0.002	4
High EC Control @ 5000 µS/cm	16.7	0.4	5	4316	303	5	9.5	0.5	5	7.87	0.06	5	19.7	-	1	0.003	0.001	4
Montezuma Slough at Nurse Slough (609)	16.4	0.3	5	4571	387	5	9.6	0.2	5	7.79	0.15	5	40.9	-	1	0.002	0.000	5
Suisun Bay off Chipps Island (508)	16.3	0.4	5	3691	173	5	9.7	0.2	5	7.83	0.13	5	15.4	-	1	0.002	0.001	5
Napa River at Riverside Blvd Terminus (340)	16.5	0.3	5	11318	997	5	9.6	0.2	5	7.76	0.14	5	14.9	-	1	0.001	0.002	5

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Table D11-1. Results of a 7-day *H. transpacificus* test initiated on 6/07/07 examining the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 6/06/07 - 6/07/07.

Treatment	Survival (%) ¹					Weight (mg) ¹		Biomass (mg) ¹			
	mean	se	A	B	C	mean	se	mean	se	A	B
Low EC Control @ 160 µS/cm	89.4	2.0	A	B		2.215	0.118	1.981	0.121	A	B
Sacramento R. at Grand Island (711)	74.8	6.1		B	C	1.952	0.153	1.471	0.192		B
Sacramento R. at Hood DWR Station	68.4	4.6			C	2.124	0.246	1.447 ²	0.171		B
Old River at Holland Cut (915)	87.1	5.5	A	B	C	1.900	0.125	1.659	0.157	A	B
Sacramento R. at Vernalis DWR Station	86.6	2.7	A	B	C	2.184	0.115	1.885	0.087	A	B
Hatchery Water @ 2500 µS/cm	97.7	2.3	A			2.182	0.127	2.124	0.075	A	B
High EC Control @ 5000 µS/cm	93.6	4.0	A	B		2.514	0.214	2.377	0.292	A	
Montezuma Slough at Nurse Slough (609)	93.6	4.0	A	B		2.206	0.127	2.071	0.180	A	B
Suisun Bay off Chipps Island (508)	93.8	2.1	A	B		2.296	0.188	2.140	0.136	A	B
Napa River at Riverside Blvd Terminus (340)	89.2	3.9	A	B		2.725	0.205	2.409	0.122	A	

Survival PMSD: 19.6%

Weight PMSD: 37.2%

Biomass PMSD: 37.3%

1. Highlighted areas indicate significant reduction in survival, weight or biomass compared to the appropriate control.

Data were analyzed using EPA standard statistical procedures, one-way ANOVAs, and Tukey's multiple comparison procedure.

2. This biomass was found to differ significantly from the control by EPA statistical procedures, but not by ANOVA.

Smelt were 44 days old at test initiation.

Table D11-2. Water chemistry at field conditions of samples collected by the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 6/06/07 - 6/07/07.

Treatment	Field Chemistry					Unionized Ammonia (mg/L)	Turbidity (NTU)
	SC (µS/cm)	EC (µS/cm)	Temp (°C)	DO (mg/L)	pH		
Sacramento R. at Grand Island (711)	205.1	185.7	20.1	7.7	7.88	0.013	3.4
Sacramento R. at Hood DWR Station	327.7	172.4	21.9	7.6	7.32	0.010	5.5
Old River at Holland Cut (915)	343.9	316.8	20.7	7.9	8.14	0.002	4.7
Sacramento R. at Vernalis DWR Station	451.9	396.8	19.6	11	8.39	0.000	5.0
Montezuma Slough at Nurse Slough (609)	5570	4946.2	19.4	8.5	7.77	0.001	18.9
Suisun Bay off Chipps Island (508)	4035	3494.3	18.3	9.5	7.9	0.002	11.5
Napa River at Riverside Blvd Terminus (340)	13870	15170	20.7	8.7	7.62	0.000	6.8

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Table D11-3. Water chemistry during a *H. transpacificus* (Delta Smelt) 7-day test initiated on 6/07/07 evaluating the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 6/06/07 - 6/07/07.

Treatment	Temp (°C)		EC (µS/cm)		DO (mg/L)		pH		Turbidity (NTU)		Un-ionized Ammonia (mg/L)		Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)
	N = 5		N = 5		N = 5		N = 5		N = 4		N = 5			
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD		
Low EC Control @ 160 µS/cm	16.6	0.6	250	106	8.8	0.5	7.66	0.22	8.7	4.1	0.006	0.005	44	34
Sacramento R. at Grand Island (711)	16.6	0.6	222	103	9.7	0.2	7.88	0.14	2.1	0.8	0.019	0.005	60	68
Sacramento R. at Hood DWR Station	16.7	0.9	217	94	9.6	0.0	7.78	0.18	2.6	0.9	0.010	0.004	64	72
Old River at Holland Cut (915)	16.8	0.7	356	101	9.8	0.2	7.88	0.12	2.4	1.0	0.004	0.002	76	70
Sacramento R. at Vernalis DWR Station	16.8	1.0	438	76	9.7	0.2	8.10	0.23	3.5	1.1	0.004	0.003	148	82
Hatchery Water @ 2500 µS/cm	16.8	0.7	2273	141	9.2	0.3	7.79	0.15	6.4	1.9	0.007	0.007	580	390
High EC Control @ 5000 µS/cm	16.7	0.8	4305	278	9.0	0.3	7.76	0.13	6.7	1.9	0.007	0.007	640	70
Montezuma Slough at Nurse Slough (609)	16.7	0.6	4644	336	9.7	0.3	7.81	0.11	7.7	2.8	0.002	0.002	3200	120
Suisun Bay off Chipps Island (508)	16.6	0.8	3538	197	9.7	0.3	7.87	0.09	6.3	1.3	0.003	0.002	944	120
Napa River at Riverside Blvd Terminus (340)	16.8	0.7	12888	1118	9.5	0.4	7.82	0.11	3.7	1.2	0.002	0.002	1744	134

Table D12-1. Results of a *H. transpacificus* 7-day test initiated on 6/22/07 evaluating the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 6/20/07 - 6/21/07. Smelt were 59 days old at test initiation.

Treatment	Survival (%) ¹	
	mean	se
Low EC Control	85.0	5.0
Sacramento R. at tip of Grand Island (711)	72.5	9.5
Sacramento R. at Hood DWR Station	80.2	5.9
Old R. at Holland Cut (915)	75.1	8.4
Hatchery Rearing Water Control	81.7	8.0
High EC Control @ 5000 uS/cm and 11 NTU	94.4	3.2
Montezuma Slough at Nurse Slough (609)	88.9	7.9
Suisun Bay off Chipps Island (508)	91.9	5.3
Napa River at Riverside Blvd Terminus (340)	94.7	3.1
Low Turbidity Control	83.3	9.6

Tukey's MSD: 33.6%

1. No significant reductions in survival were seen in any sample, and no significant differences in survival were seen among the various controls. Data were analyzed using both Tukey's multiple comparison procedure and EPA standard statistical protocols.

Table D12-2. Water chemistry at field conditions of samples collected by the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 6/20/07 - 6/21/07.

Sample	Field Chemistry				
	SC (μ S/cm)	EC (μ S/cm)	Temp ($^{\circ}$ C)	DO (mg/L)	pH
Sacramento R. at Grand Island (711)	172.6	161.9	21.9	8.1	7.80
Sacramento R. at Hood DWR Station	124.1	119.4	23.1	7.0	7.32
Old River at Holland Cut (915)	291.5	287.4	24.3	7.8	8.06
Montezuma Slough at Nurse Slough (609)	7000	6412	20.8	7.9	7.84
Suisun Bay off Chipps Island (508)	5360	4931	21.0	8.8	7.97
Napa River at Riverside Blvd Terminus (340)	17020	17088	25.2	8.7	7.76

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Table D12-3. Water chemistry during a *H. transpacificus* (Delta Smelt) 7-day test initiated on 6/22/07 evaluating the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 6/20/07 - 6/21/07.

Treatment	Temp (°C)			EC (µS/cm)			DO (mg/L)			pH			Turbidity (NTU)			Un-ionized Ammonia (mg/L)			Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)
	Mean	SD	N	Mean	SD	N	Mean	SD	N	Mean	SD	N	Mean	SD	N	Mean	SD	N		
Low EC Control	17.2	0.9	8	164	19	8	9.2	0.5	8	7.78	0.19	8	7.8	0.7	7	0.000	0.015	5	36	26
Sacramento R. at tip of Grand Island (711)	16.9	1.0	8	171	24	8	9.5	0.3	8	7.96	0.08	8	4.0	1.3	8	0.001	0.012	5	64	62
Sacramento R. at Hood DWR Station	16.8	0.6	8	134	26	8	9.5	0.3	8	7.98	0.13	8	2.5	1.6	8	0.003	0.009	5	52	50
Old R. at Holland Cut (915)	16.7	1.2	8	273	23	8	9.5	0.3	8	8.03	0.12	8	2.3	0.8	8	0.001	0.008	5	80	66
Hatchery Rearing Water Control	17.1	0.5	8	924	19	8	9.4	0.3	8	7.98	0.08	8	5.7	0.8	7	0.004	0.010	4	144	86
High EC Control	17.2	0.6	8	4146	145	8	9.4	0.4	8	7.89	0.09	8	7.0	1.4	7	0.002	0.010	4	640	70
Montezuma Slough at Nurse Slough (609)	16.7	1.3	8	5040	542	8	9.4	0.3	8	7.93	0.15	8	8.9	5.8	8	0.001	0.006	5	820	70
Suisun Bay off Chipps Island (508)	16.7	1.0	8	4040	370	8	9.5	0.4	8	7.93	0.10	8	7.8	6.6	8	0.001	0.006	5	500	76
Napa River at Riverside Blvd Terminus (340)	17.1	0.8	8	14129	1141	8	9.3	0.4	8	7.92	0.11	8	3.5	1.3	8	0.000	0.003	5	2000	141
Low Turbidity Control	16.9	1.5	8	948	54	8	9.3	0.7	8	7.95	0.09	8	1.8	0.5	7	0.003	0.005	4	160	82

Table D13-1. Results of a *H. transpacificus* 96-hour test initiated on 7/26/07 evaluating the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 7/25/07 - 7/26/07.

Treatment	Survival (%) ¹	
	mean	se
Low EC Control	61.0	8.4
Sacramento R. at tip of Grand Island (711)	29.3	6.3
Sacramento R. at Hood DWR Station	56.5	14.9
Old R. at Holland Cut (915)	51.9	5.3
San Joaquin R. at Vernalis	66.1	11.5
Hatchery Rearing Water Control	40.8	8.9
High EC Control	26.5	6.2
Montezuma Slough at Nurse Slough (609)	41.3	10.9
Suisun Bay off Chipps Island (508)	32.9	5.3
Napa River at Riverside Blvd Terminus (340)	57.5	11.3
Low Turbidity Control ²	24.6	3.1

1. Data were analyzed using both EPA standard statistical protocols and Tukey's multiple comparison procedure. Highlighted areas indicate significant reduction in survival or biomass compared to the appropriate conductivity control when analyzed by EPA statistics. Tukey's multiple comparison procedure was unable to detect any significant differences.

2. The low turbidity control was not found to be significantly different than the hatchery water control.

Smelt were 54 days old at test initiation.

Table D13-2. Water chemistry at field conditions of samples collected by the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 7/25/07 - 7/26/07.

Sample	Field Chemistry				
	SC (μ S/cm)	EC (μ S/cm)	Temp ($^{\circ}$ C)	DO (mg/L)	pH
Sacramento R. at Grand Island (711)	141.5	134.7	22.6	8.1	7.29
Sacramento R. at Hood DWR Station	154.6	148.1	22.9	8.1	7.64
Old River at Holland Cut (915)	328.4	320.5	23.8	8.0	7.39
San Joaquin R. at Vernalis	587	583.5	24.7	9.7	8.43
Montezuma Slough at Nurse Slough (609)	8150	7808	22.9	8.2	7.73
Suisun Bay off Chipps Island (508)	5030	4789	22.6	8.4	7.89
Napa River at Riverside Blvd Terminus (340)	24400	23814	23.8	6.6	7.64

Table D13-3. Water chemistry during a *H. transpacificus* (Delta Smelt) 96-hour test initiated on 7/26/07 evaluating the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 7/25/07 - 7/26/07.

Treatment	Temp (°C)		EC (µS/cm)		DO (mg/L)		pH		Turbidity (NTU)		Un-ionized Ammonia (mg/L)		Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)
	N = 5		N = 5		N = 5		N = 5		N = 4		N = 4			
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD		
Low EC Control	16.6	0.7	219	75	9.6	0.2	7.69	0.27	5.5	0.7	0.007	0.006	-	-
Sacramento R. at tip of Grand Island (711)	16.5	0.6	179	64	9.8	0.1	7.84	0.17	3.1	0.4	0.011	0.003	52	57
Sacramento R. at Hood DWR Station	16.9	0.9	200	111	9.7	0.1	7.90	0.18	3.1	0.3	0.004	0.003	56	60
Old R. at Holland Cut (915)	16.7	0.8	325	46	9.7	0.2	7.91	0.18	2.7	0.6	0.005	0.002	72	59
San Joaquin R. at Vernalis	16.4	0.8	523	42	10.0	0.3	8.19	0.16	5.2	1.2	0.009	0.001	136	105
Hatchery Rearing Water Control	16.6	0.8	1746	256	9.8	0.3	7.88	0.10	2.2	0.7	0.006	0.004	-	-
High EC Control	16.6	0.6	4085	250	9.6	0.3	7.87	0.13	1.6	0.8	0.004	0.003	-	-
Montezuma Slough at Nurse Slough (609)	16.5	0.5	6558	372	9.8	0.2	7.88	0.13	4.4	0.9	0.004	0.003	760	95
Suisun Bay off Chipps Island (508)	16.6	0.6	4243	158	9.7	0.2	7.85	0.17	3.1	0.6	0.004	0.003	532	66
Napa River at Riverside Blvd Terminus (340)	16.6	0.8	19248	1456	9.4	0.3	7.79	0.19	4.3	0.4	0.003	0.002	3080	139
Low Turbidity Control	16.7	0.8	1133	69	9.7	0.2	7.69	0.30	1.3	0.2	0.001	0.002	-	-

Table D14-1. Results of a *H. transpacificus* 7-day test initiated on 8/09/07 evaluating the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 8/8/07 - 8/09/07.

Treatment	Survival (%) ¹	
	mean	se
Low EC Control @ 135 μ S/cm	82.3	7.3
Sacramento R. at tip of Grand Island (711)	58.9	9.9
Sacramento R. at Hood DWR Station	62.5	17.5
Old R. at mouth of Holland Cut (915)	75.0	5.0
San Joaquin R. at Vernalis DWR Station	96.9	3.1
Hatchery Water @ 1746 μ S/cm	92.2	4.8
High EC Control @ 5000 μ S/cm	93.7	3.7
Montezuma Slough at Nurse Slough (609)	90.5	3.2
Suisun Bay off Chipps Island (508)	92.9	4.1
Napa R. at Riverside Blvd terminus (340)	81.5	8.9
Low Turbidity Control (2 - 3 NTU) at 3131 μ S/cm	70.8	13.1

1. Highlighted areas indicate significant reduction in survival compared to the appropriate control. Data were analyzed using both USEPA standard statistical protocols and Tukey's multiple comparison procedure.

Smelt were 92 days old at test initiation.

Table D14-2. Water chemistry at field conditions of samples collected by the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 8/8/07 - 8/09/07.

Sample	Field Chemistry				
	SC (μ S/cm)	EC (μ S/cm)	Temp (°C)	DO (mg/L)	pH
Sacramento R. at tip of Grand Island (711)	142.6	134.3	22.1	8.6	7.86
Sacramento R. at Hood DWR Station	153.5	140.6	20.8	8.3	7.22
Old R. at mouth of Holland Cut (915)	396.6	379.1	22.8	8.2	7.65
San Joaquin R. at Vernalis DWR Station	-	-	-	-	-
Montezuma Slough at Nurse Slough (609)	8970	8127	20.3	8.3	7.43
Suisun Bay off Chipps Island (508)	7140	6497	20.5	8.7	7.88
Napa R. at Riverside Blvd terminus (340)	25760	23905	21.4	7.0	7.25

Table D14-3. Water chemistry during a *H. transpacificus* (Delta Smelt) 7-day test initiated on 8/09/07 evaluating the toxicity of samples collected by the UC Davis Aquatic Toxicology Laboratory (UCDATL) and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) on 8/8/07 - 8/09/07.

Treatment	Temp (°C)		EC (µS/cm)		DO (mg/L)		pH		Turbidity (NTU)		Un-ionized Ammonia (mg/L)			Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)
	N = 8		N = 8		N = 8		N = 8		N = 7		Mean	SD	N		
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	N		
Low EC Control	17.1	0.3	204	86	9.3	0.1	7.46	0.11	4.2	0.9	0.003	0.001	4	28	24
Sacramento R. at tip of Grand Island (711)	17.1	0.4	179	72	9.4	0.2	7.74	0.09	2.1	0.5	0.005	0.002	5	52	58
Sacramento R. at Hood DWR Station	17.2	0.5	189	88	9.6	0.2	7.80	0.08	2.7	0.6	0.003	0.001	5	56	62
Old R. at mouth of Holland Cut (915)	17.2	0.6	363	77	9.5	0.3	7.86	0.06	2.3	0.5	0.004	0.002	5	108	58
San Joaquin R. at Vernalis DWR Station	17.1	0.4	564	54	9.5	0.2	8.08	0.06	4.5	0.7	0.007	0.003	5	152	112
Hatchery Water Control	17.2	0.4	1602	89	8.7	0.2	7.62	0.11	5.3	0.5	0.004	0.001	4	208	60
High EC Control	17.4	0.5	4103	77	8.9	0.3	7.65	0.09	5.5	0.8	0.003	0.001	4	550	71
Montezuma Slough at Nurse Slough (609)	17.3	0.6	7220	266	9.4	0.3	7.89	0.07	4.9	1.9	0.002	0.001	5	1020	110
Suisun Bay off Chipps Island (508)	17.1	0.7	4852	1997	9.5	0.2	7.77	0.07	3.3	2.5	0.002	0.001	5	152	70
Napa R. at Riverside Blvd terminus (340)	16.9	0.6	19993	1427	9.0	0.4	7.79	0.08	3.4	1.2	0.002	0.001	5	3160	180
Low Turbidity Control	17.3	0.5	1497	48	9.5	0.3	6.97	1.09	1.1	0.5	0.001	0.000	4	126	24

Reference Toxicant Dilution Series – Copper

Table D15-1. Summary of a 7-day *H. transpacificus* test initiated on 9/01/05 examining the toxicity of Copper (Cu⁺).

Treatment	Rep	# Surviving Fish by Test Day						
		1	2	3	4	5	6	7
0 ppb Control (Dilute Well Water)	1	5	4	4	4	4	4	4
	2	5	5	5	4	4	4	4
50 ppb Cu ⁺	1	4	3	3	2	2	2	2
	2	0	0	0	0	0	0	0
200 ppb Cu ⁺	1	0	0	0	0	0	0	0
	2	0	0	0	0	0	0	0
500 ppb Cu ⁺	1	0	0	0	0	0	0	0
	2	0	0	0	0	0	0	0
1000 ppb Cu ⁺	1	0	0	0	0	0	0	0
	2	0	0	0	0	0	0	0

Table D15-2. Summary of water chemistry measurements taken during a 7-day *H. transpacificus* test initiated on 9/01/05 examining the toxicity of Copper (Cu⁺).

Treatment	Temp (°C)	pH	EC (uS/cm)	DO (mg/L)	Ammonia Nitrogen (mg/L)
Lab Control (Dilute Well Water)	20	8.52	507	9.2	0.15
50 ppb Cu ⁺	20	8.46	440	9.1	0.09
200 ppb Cu ⁺	21	8.43	266	8.7	0.10
500 ppb Cu ⁺	21	8.46	235	9.0	0.08
1000 ppb Cu ⁺	21	8.46	325	9.1	0.10

Table D16-1. Summary of 7-day *H. transpacificus* Cu⁺ reference toxicant test conducted using dilute well water spiked with copper chloride.¹

Treatment	Survival (%) ²		Length (cm) ²		Weight (g) ²	
	mean	se	mean	se	mean	se
Laboratory Control (Dilute Well Water)	100 ^P	0.0	3.42	0.04	0.24	0.00
5 ppb Cu ⁺	93	6.7	3.53	0.05	0.24	0.01
10 ppb Cu ⁺	95	2.9	3.49	0.04	0.23	0.00
25 ppb Cu ⁺	40	4.1	3.57	0.11	0.26	0.02
50 ppb Cu ⁺	23	4.7	3.52	0.08	0.26	0.02

P. The laboratory control met the criteria for test acceptability.

1. This test was set up on 9/14/05.

2. Highlighted areas indicate a significant reduction in survival, length, or weight when compared to the laboratory control. All endpoints were analyzed according to EPA standard methods. (1-tailed test, P < 0.05)

	Survival	
	96 hr	7 day
NOEC	10	10
LOEC	25	25
LC50	33.5	24.7

Table D16-2. Summary of water chemistry measurements taken during a 7-day *H. transpacificus* reference toxicant test initiated on 9/14/05 using dilute well water spiked with copper chloride.¹

Treatment	Temp (°C)	Lab pH	EC (uS/cm)	DO (mg/L)	Ammonia Nitrogen (mg/L)
Lab. Control (Dilute Well Water)	21	8.40	431	8.8	0.28
5 ppb Cu ⁺	21	8.49	456	8.7	0.24
10 ppb Cu ⁺	21	8.48	461	9.0	0.23
25 ppb Cu ⁺	21	8.46	455	8.8	0.37
50 ppb Cu ⁺	21	8.39	457	8.9	0.14

Esfenvalerate Exposures

Table D17. Results of a *H. transpacificus* 24-hour test initiated 5/23/06 evaluating the toxicity of esfenvalerate to 10 day old fish.

Treatment	4-hour Survival (%) ¹		24-hour Survival (%) ¹		4-hour Normal Swimming (%) ¹		24-hour Normal Swimming (%) ¹	
	mean	se	mean	se	mean	se	mean	se
Method Control	97.5	2.5	77.5	4.8	92.5	2.5	62.5	2.5
Solvent Control	97.5	2.5	82.5	2.5	93.0	4.4	72.5	2.5
0.03125 µg/L Esfenvalerate	100.0	0.0	87.5	9.5	97.7	2.3	65.0	11.9
0.0625 µg/L Esfenvalerate	97.5	2.5	75.0	9.6	95.0	2.9	27.5	10.3
0.125 µg/L Esfenvalerate	97.5	2.5	57.5	7.5	87.5	4.8	0.0	0.0
0.25 µg/L Esfenvalerate	100.0	0.0	25.0	2.9	77.5	2.5	0.0	0.0
0.5 µg/L Esfenvalerate	100.0	0.0	0.0	0.0	60.0	5.8	0.0	0.0

1. Highlighted areas indicate significant reduction in survival or biomass compared to the solvent control. Data were analyzed using USEPA standard statistical protocols.

Table D18. Results of a *H. transpacificus* 24-hour test initiated 6/01/06 evaluating the toxicity of esfenvalerate to 31 day old fish.

Treatment	4-hour Survival (%) ¹		24-hour Survival (%) ¹		4-hour Normal Swimming (%) ¹		24-hour Normal Swimming (%) ¹	
	mean	se	mean	se	mean	se	mean	se
Method Control	100.0	0.0	100.0	0.0	100.0	0.0	100.0	0.0
Solvent Control	100.0	0.0	100.0	0.0	100.0	0.0	97.5	2.5
0.03125 µg/L Esfenvalerate	100.0	0.0	100.0	0.0	97.5	2.5	97.5	2.5
0.0625 µg/L Esfenvalerate	100.0	0.0	100.0	0.0	100.0	0.0	87.5	4.8
0.125 µg/L Esfenvalerate	100.0	0.0	95.0	2.9	92.5	2.5	70.0	4.1
0.25 µg/L Esfenvalerate	100.0	0.0	87.5	12.5	95.0	5.0	37.5	13.1
0.5 µg/L Esfenvalerate	100.0	0.0	55.0	9.6	87.5	4.8	0.0	0.0
1.0 µg/L Esfenvalerate	100.0	0.0	10.0	7.1	75.0	2.9	0.0	0.0

1. Highlighted areas indicate significant reduction in survival or biomass compared to the solvent control. Data were analyzed using USEPA standard statistical protocols.

Table D19. Results of a *H. transpacificus* 24-hour test initiated 5/23/06 evaluating the toxicity of esfenvalerate to 52 day old fish.

Treatment	4-hour Survival (%) ¹		24-hour Survival (%) ¹		4-hour Normal Swimming (%) ¹		24-hour Normal Swimming (%) ¹	
	mean	se	mean	se	mean	se	mean	se
Method Control	100.0	0.0	100.0	0.0	82.5	4.8	82.5	4.8
Solvent Control	100.0	0.0	97.5	2.5	72.5	2.5	70.0	0.0
0.03125 µg/L Esfenvalerate	100.0	0.0	100.0	0.0	80.0	0.0	80.0	0.0
0.0625 µg/L Esfenvalerate	100.0	0.0	92.5	2.5	72.5	6.3	65.0	5.0
0.125 µg/L Esfenvalerate	100.0	0.0	95.0	5.0	57.5	4.8	52.5	6.3
0.25 µg/L Esfenvalerate	87.5	4.8	40.0	12.2	47.5	13.1	0.0	0.0

1. Highlighted areas indicate significant reduction in survival or biomass compared to the solvent control. Data were analyzed using USEPA standard statistical protocols.

Table D20. Results of a *H. transpacificus* 24-hour test initiated 10/18/06 evaluating the toxicity of esfenvalerate to 204 day old fish.

Treatment	4-hour Survival (%) ¹		24-hour Survival (%) ¹		4-hour Normal Swimming (%) ¹		24-hour Normal Swimming (%) ¹	
	mean	se	mean	se	mean	se	mean	se
Method Control	100.0	0.0	100.0	0.0	100.0	0.0	92.5	2.5
Solvent Control	100.0	0.0	95.0	2.9	100.0	0.0	92.5	2.5
0.1 µg/L Esfenvalerate	100.0	0.0	94.7	3.1	100.0	0.0	82.2	7.4
0.25 µg/L Esfenvalerate	100.0	0.0	87.5	2.5	97.5	2.5	75.0	6.5
0.5 µg/L Esfenvalerate	100.0	0.0	82.5	6.3	100.0	0.0	37.5	2.5
1.0 µg/L Esfenvalerate	100.0	0.0	20.0	10.0	92.5	4.8	0.0	0.0
5.0 µg/L Esfenvalerate	100.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0

1. Highlighted areas indicate significant reduction in survival or biomass compared to the solvent control. Data were analyzed using USEPA standard statistical protocols.

Treatment	Length (mm) ¹		Weight (g) ¹	
	mean	se	mean	se
Method Control	61.2	2.2	1.61	0.17
Solvent Control	59.7	1.1	1.50	0.09
0.1 µg/L Esfenvalerate	60.3	0.9	1.52	0.08
0.25 µg/L Esfenvalerate	60.9	0.6	1.60	0.08
0.5 µg/L Esfenvalerate	61.1	1.6	1.62	0.17
1.0 µg/L Esfenvalerate	54.9	5.1	1.18	0.28
5.0 µg/L Esfenvalerate	-	-	-	-

Appendix E

Pimephales promelas

(Fathead Minnow)

Table E1-1. Results of a *P. promelas* (Fathead Minnow) 7-day test initiated on 5/23/07 evaluating the toxicity of water collected by UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) from the San Joaquin River at the Stockton WWTP on 5/22/07.

Treatment	Survival (%) ¹		Biomass (mg) ¹	
	mean	se	mean	se
DIEPAMH	100.0	0.0	0.386	0.011
SJR @ WWTP (Stockton)	97.5	2.5	0.429	0.006

1. No significant reductions in survival or biomass were seen compared to the DIEPAMH control.

Table E1-2. Water chemistry during a *P. promelas* (Fathead Minnow) 7-day test initiated on 5/23/07 evaluating the toxicity of water collected by UC Davis Aquatic Toxicology Laboratory and the California Department of Fish and Game (CDFG) for the Department of Water Resources (DWR) from the San Joaquin River at the Stockton WWTP on 5/22/07.

Treatment	Field Chemistry				Unionized Ammonia (mg/L)	Turbidity (NTU)
	SC (uS/cm)	Temp (°C)	DO (mg/L)	pH		
San Joaquin River at Stockton WW	345.3	20.3	11	8.8	0.016	31

Treatment	Laboratory Chemistry						Hardness (mg/L as CaCO ₃)	Alkalinity (mg/L as CaCO ₃)	
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH			Max pH
DIEPAMH	286	24	25	7.1	8.4	7.66	8.18	92	60
San Joaquin River at Stockton WW	322	24	25	7.8	8.2	7.81	8.24	80	60

Appendix F
Quality Assurance/Quality Control

Table F1. Frequency of QA/QC samples sharing equivalent results: 2006-2007

Quality Assurance Samples	<i>H. azteca</i> Survival		<i>H. azteca</i> Weight	
	Sample Size	% Agreement	Sample Size	% Agreement
Field Duplicates	39	100	39	100
Bottle Blanks	16	100	16	94
Trip Blanks	13	100	13	100

Table F2. Individual RPDs of water chemistry measurements on duplicate samples collected February – June 2006.

Field Duplicate & Sample Date	Water Chemistry Measurements					
	EC	DO	pH	Hardness	Alkalinity	Ammonia
Site 915 February 21, 2006	2.29 8.55	4.55 5.33 0.00	1.58 0.53	0.00	3.57	200.00 ^{A,B}
Site 804 March 7, 2006	43.30 ^B 1.62	2.33 2.78 14.59	2.85 1.44 6.94	5.13	0.00	11.76
Site 915 March 20, 2006	71.28 ^B 5.94	1.24 6.06 3.92	1.38 0.26 0.52	24.39	25.45	168.00 ^{A,B}
Site 915 April 3, 2006	16.17 23.33	5.06 2.78 7.50	0.85 1.83 3.87	23.53	50.00 ^B	111.11 ^{A,B}
Site 340 April 4, 2006	47.64 ^B 3.56	1.24 1.44 8.00	1.11 0.13 3.74	22.22	0.00	18.18
Site 910 April 17, 2006	10.58 5.58	2.30 1.38 0.00	1.90 1.01 0.26	6.45	0.00	8.00
Site Light 55 May 1, 2006	0.23 0.82	3.55 1.26 4.38	1.19 0.63 0.25	5.56	1.63	40.00 ^{A,B}
Site 812 May 2, 2006	8.70 2.86	0.00 0.00 4.58	0.13 0.39 0.26	8.00	4.65	46.15 ^{A,B}
Site Light 55 May 15, 2006	4.52 1.88	1.24 2.74 0.00	0.00 7.02 0.12	2.41	1.44	0.00
Site 804 June 1, 2006	3.98 18.20	1.20 3.57 0.00	0.88 0.14 0.26	0.00	13.33	28.57
Site 804 June 13, 2006	3.13 0.23	2.35 4.80 1.87	1.40 1.06	9.52	2.47	0.00
Site 915 June 28, 2006	1.81 1.89	1.17 0.00 0.00	1.13 5.15 0.80	24.39	2.74	0.00

Table F3. Average RPDs of water chemistry measurements on duplicate samples collected February – June 2006.

Field Duplicate & Sample Date	Water Chemistry Measurements								
	EC			DO			pH		
	Sample Size	Average	Standard Deviation	Sample Size	Average	Standard Deviation	Sample Size	Average	Standard Deviation
Site 915 February 21, 2006	2	5.42	4.43	3	3.29	2.88	2	1.05	0.74
Site 804 March 7, 2006	2	22.46	29.47	3	6.57	6.96	3	3.75	2.86
Site 915 March 20, 2006	2	38.61	46.20	3	3.74	2.41	3	2.41	0.58
Site 915 April 3, 2006	2	19.75	5.07	3	5.11	2.36	3	2.18	1.54
Site 340 April 4, 2006	2	25.60	31.17	3	3.56	3.85	3	1.66	1.87
Site 910 April 17, 2006	2	8.08	3.54	3	1.23	1.16	3	1.06	0.82
Site Light 55 May 1, 2006	2	0.52	0.42	3	3.06	1.62	3	0.69	0.47
Site 812 May 2, 2006	2	5.78	4.13	3	1.53	2.64	3	0.26	0.13
Site Light 55 May 15, 2006	2	3.20	1.87	3	1.33	1.37	3	2.38	4.02
Site 804 June 1, 2006	2	11.09	10.05	3	1.59	1.59	3	0.43	0.40
Site 804 June 13, 2006	2	1.68	2.05	3	3.01	1.57	2	1.23	0.12
Site 915 June 28, 2006	2	1.85	0.05	3	0.39	0.68	3	2.36	2.42

Table F4. Individual RPDs of water chemistry measurements on duplicate samples collected July 2006 – January 2007.

Field Duplicate & Sample Date	Water Chemistry Measurements					
	EC	DO	pH	Hardness	Alkalinity	Ammonia
Site 915	1.74	1.23	0.37			
July 25, 2006	0.95	1.21	3.05	7.41	14.81	200.00 ^{A,B}
		1.74				
Site 405	0.47	1.24	1.16			
August 9, 2006	196.04 ^B	2.41	0.81	63.59 ^B	9.52	200.00 ^{A,B}
		3.17				
Site 609	0.40	0.00	0.50			
August 23, 2006	0.39	0.00	0.52	7.19	0.00	15.38
		2.74				
Site Light 55	2.41	4.76	0.37			
September 5, 2006	0.06	1.21	0.38	3.92	3.35	200.00 ^{A,B}
		2.94				
Site 812	46.65 ^B	1.26	1.49			
September 20, 2006	43.36 ^B	0.00	0.25	40.00 ^B	21.12	0.00
		6.90				
Site 340	1.23	2.41	1.18			
October 5, 2006	0.55	1.13	0.50	0.36	1.42	0.00
		2.60				
Site 602	0.89	4.60	1.40			
October 18, 2006	4.23	1.20	3.05	2.35	13.33	24.00
		1.26				
Site 711	22.10	2.35	1.64			
October 31, 2006	0.73	2.33	0.89	14.29	0.00	20.69
		1.32				
Site Light 55	59.21 ^B	0.00	0.12			
January 4, 2007	7.17	1.18	0.25	8.70	0.00	0.00
		1.40				

Table F5. Average RPDs of water chemistry measurements on duplicate samples collected July 2006 – January 2007.

Field Duplicate & Sample Date	Water Chemistry Measurements								
	EC			DO			pH		
	Sample Size	Average	Standard Deviation	Sample Size	Average	Standard Deviation	Sample Size	Average	Standard Deviation
Site 915 July 25, 2006	4	1.35	0.56	6	1.39	0.30	4	1.71	1.89
Site 405 August 9, 2006	4	98.25	138.29	6	2.28	0.97	4	0.98	0.25
Site 609 August 23, 2006	4	0.40	0.00	6	0.92	1.58	4	0.51	0.02
Site Light 55 September 5, 2006	4	1.23	1.66	6	2.97	1.77	4	0.38	0.01
Site 812 September 20, 2006	4	45.01	2.33	6	22.45	31.96	4	0.87	0.88
Site 340 October 5, 2006	4	0.89	0.48	6	2.05	0.80	4	0.84	0.48
Site 602 October 18, 2006	4	2.56	2.36	6	2.35	1.95	4	2.22	1.16
Site 711 October 31, 2006	4	11.41	15.11	6	2.00	0.59	4	1.27	0.53
Site Light 55 January 4, 2007	4	33.19	36.80	6	0.86	0.75	4	0.19	0.10

Table F6. Individual RPDs of water chemistry measurements on duplicate samples collected February – August 2007.

Field Duplicate & Sample Date	Water Chemistry Measurements					
	EC	DO	pH	Hardness	Alkalinity	Ammonia
Site 704 March 16, 2007	2.00 3.03	1.43 4.76 1.29	0.13 0.13	5.41	32.65	8.00
Site 902 July 26, 2007	1.13 1.05	2.35 1.16 1.40	0.51 0.53	11.11	0.00	560.00 ^{A,B}
Site 504 August 8, 2007	2.30 3.74	3.73 4.60 1.57	0.51 0.13	13.79	0.00	40.00 ^{A,B}
Site 804 August 22, 2007	1.54 2.74	1.18 6.98 2.99	0.25 0.90	1.87	0.00	0.00
Site 910 March 29, 2007	4.41 0.00	1.20 1.16 1.44	0.38 0.26	2.74	0.00	0.00
Site 602 April 11, 2007	1.82 4.20	2.30 1.20 7.52	0.76 0.80	70.06 ^B	0.00	15.38
Site 804 April 25, 2007	3.00 1.23	5.92 8.19 0.00	1.38 0.40	12.24	0.00	6.90
Site 504 May 10, 2007	0.62 0.38	1.24 2.35 2.86	0.13 0.26	3.62	10.53	11.76
Site 812 May 24, 2007	4.64 0.46	1.21 2.53 2.78	0.13 0.26	0.00	3.17	22.22
Site 711 June 20, 2007	0.43 3.33	1.21 1.14 1.31	0.75 0.64	6.06	17.65	28.57

Table F7. Average RPDs of water chemistry measurements on duplicate samples collected February – August 2007.

Field Duplicate & Sample Date	Water Chemistry Measurements								
	EC			DO			pH		
	Sample Size	Average	Standard Deviation	Sample Size	Average	Standard Deviation	Sample Size	Average	Standard Deviation
Site 704 March 16, 2007	4	2.51	0.73	6	2.40	2.05	4	0.13	0.00
Site 910 March 29, 2007	4	2.20	3.12	6	1.26	0.15	4	0.32	0.08
Site 602 April 11, 2007	4	3.01	1.68	6	3.67	3.38	4	0.78	0.02
Site 804 April 25, 2007	4	2.10	1.23	6	4.70	4.23	4	0.88	0.71
Site 504 May 10, 2007	4	0.50	0.17	6	2.15	0.83	4	0.20	0.10
Site 812 May 24, 2007	4	2.55	2.95	6	2.17	0.84	4	0.19	0.10
Site 711 June 20, 2007	4	1.88	2.05	6	1.22	0.08	4	0.70	0.08
Site 902 July 26, 2007	4	1.09	0.06	6	1.64	0.63	4	0.52	0.01
Site 504 August 8, 2007	4	3.02	1.04	6	3.30	1.56	4	0.32	0.26
Site 804 August 22, 2007	4	2.14	0.85	6	3.72	2.96	4	0.58	0.46

Table F8. Individual RPDs of water chemistry measurements on duplicate samples collected September – December 2007.

Field Duplicate & Sample Date	Water Chemistry Measurements					
	EC	DO	pH	Hardness	Alkalinity	Ammonia
Site Hood September 5, 2007	0.97 2.68	0.00 5.92 11.97	1.27 1.21	6.45	2.90	10.53
Site 711 October 3, 2007	0.53 1.28	1.17 8.28 1.46	0.63 0.26	3.64	1.57	0.00
Site Hood December 11, 2007	0.99 1.33	1.20 0.00 1.32	0.87 0.13	9.52	0.00	3.39
Site 910 December 12, 2007	3.59 0.87	2.38 0.00 2.56	0.37 0.25	16.67	4.88	11.76
Site 902 December 12, 2007	2.63 2.61	4.76 4.60 1.31	0.12 0.26	14.81	0.00	28.57
Site 609 December 13, 2007	2.67 1.21	4.76 0.00 2.63	0.64 0.37	0.55	2.41	4.26

Table F9. Average RPDs of water chemistry measurements on duplicate samples collected September – December 2007.

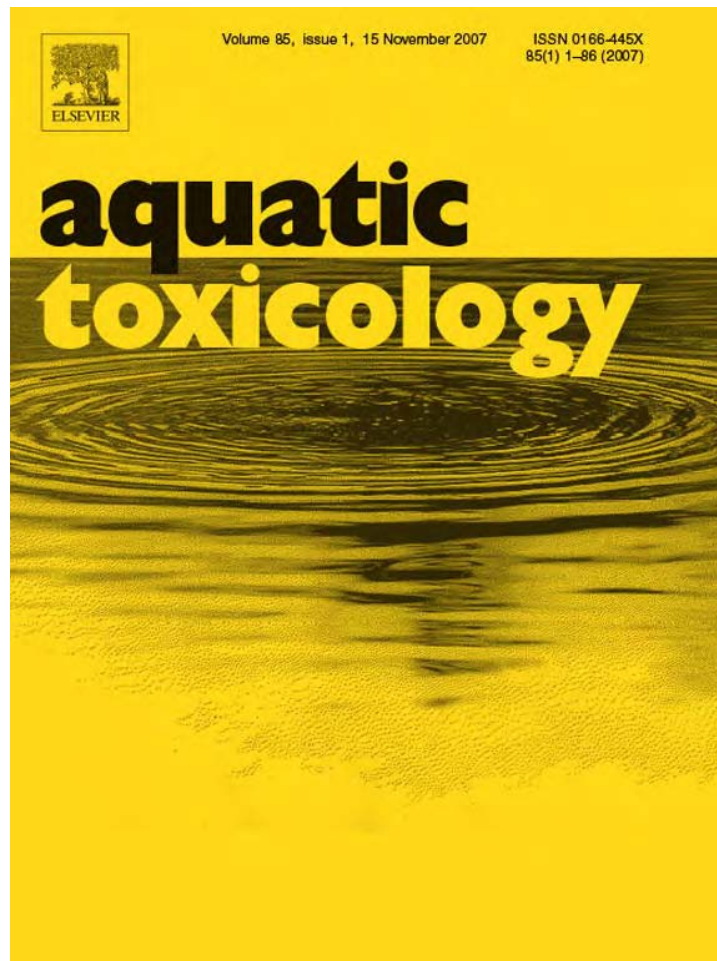
Field Duplicate & Sample Date	Water Chemistry Measurements								
	EC			DO			pH		
	Sample Size	Average	Standard Deviation	Sample Size	Average	Standard Deviation	Sample Size	Average	Standard Deviation
Site Hood September 5, 2007	4	1.82	1.21	6	5.96	5.98	4	1.24	0.04
Site 711 October 3, 2007	4	0.91	0.53	6	3.64	4.03	4	0.45	0.26
Site Hood December 11, 2007	4	1.16	0.24	6	0.84	0.73	4	0.50	0.52
Site 910 December 12, 2007	4	2.23	1.92	6	1.65	1.43	4	0.31	0.08
Site 902 December 12, 2007	4	2.62	0.01	6	3.56	1.95	4	0.19	0.09
Site 609 December 13, 2007	4	1.94	1.04	6	2.46	2.39	4	0.51	0.19

^A: Caution should be applied when interpreting water quality precision data. Although the difference between ammonia replicates is large, it is because low concentrations of ammonia were measured rather than lack of precision. For instance, comparing ammonia measurements of 0.004 vs 0.000 will yield a RPD of 200%

^B: Exceeds SWAMP RPD range

Appendix G

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Comparisons of tissue-specific transcription of stress response genes with whole animal endpoints of adverse effect in striped bass (*Morone saxatilis*) following treatment with copper and esfenvalerate

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Abstract

Changes in the gene transcription of stress response genes in resident fish can be powerful biomarkers for the identification of sublethal impacts of environmental stressors on aquatic ecosystems. In this study, we tested the effects of two reference toxicants, copper (Cu) and the pyrethroid insecticide esfenvalerate [(*S*)- α -cyano-3-phenoxybenzyl-(*S*)-2-(4-chlorophenyl)-3-methylbutyrate], on lethal (mortality) and sublethal endpoints (growth, swimming behavior, transcription levels of stress response genes) in juvenile (81–90-day-old) striped bass (*Morone saxatilis*). We established cellular stress response markers for proteotoxicity (HSP70, HSP90), phase I detoxification mechanism (CYP1A1), metal-binding (metallothionein), as well as immune-function and pathogen-defense (TGF- β , Mx-protein, nRAMP). Quantitative real-time TaqMan[®] PCR was used to examine tissue-specific changes in the transcriptome of liver, spleen, white muscle, anterior kidney and gills after 7-day Cu exposures and 24-h esfenvalerate exposures. On the transcriptome level, exposure to Cu showed strongest effects on the transcription of metallothionein in spleen tissue, causing a 4-fold increase of mRNA at 42 ppb total Cu and a 10-fold increase at 160 ppb Cu. Exposure to Cu also caused significant tissue-specific changes in gene transcription for immune-system related genes. Esfenvalerate exposure had tissue-specific effects on the transcription of HSP70, HSP90 and CYP1A1. The most significant effects were detected in liver tissue after exposure to 0.64 μ g/L esfenvalerate.

Our results show that the stress response at the transcriptome level is a more sensitive indicator for Cu and esfenvalerate exposures at low concentrations than swimming behavior, growth or mortality. The accuracy of studies on quantitative changes in the transcriptome can benefit from an initial evaluation or the inclusion of several different tissues and the use of multiple housekeeping genes.

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1. Introduction

The current rate of biodiversity loss in aquatic ecosystems threatens to disrupt their functioning in many parts of the world. In most cases a multitude of factors, including overfishing, loss and degradation of habitat, the invasion of exotic species, flow modification, and pollution all interact and simultaneously contribute to the decline of species (Bennett and Moyle, 1996).

Ecological forensic methods try to separate and identify factors for species declines, but for non-model organisms, such attempts are often difficult to realize.

In the last several years, abundance indices of numerous pelagic fish species residing in the Sacramento-San Joaquin Delta of California, USA, have shown marked declines and record lows for the endemic delta smelt (*Hypomesus transpacificus*), age-0 striped bass (*Morone saxatilis*), longfin smelt (*Spirinchus thaleichthys*) and threadfin shad (*Dorosoma petenense*) (Stevens and Miller, 1983; Stevens et al., 1985; Moyle et al., 1992; Moyle and Williams, 1990). While several of these species – in particular longfin smelt and juvenile striped bass – have shown evidence of long-term declines, there appears to have been a precipitous “step-change” to very low abundance

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during the period 2002–2004 (Bryant and Souza, 2004; Hieb et al., 2005; Feyrer et al., 2007). At present, it is unclear what might have caused this critical population decline, but toxic contaminants may be one of several factors acting individually or in concert to lower pelagic productivity.

Agricultural, industrial, urban and mining sources release contaminants into waterways, and water quality assessment studies indicate that the criteria for the protection of freshwater aquatic life have been exceeded in many Central Valley water bodies (Domagalski et al., 2000; Dubrovsky et al., 1998; De Vlaming et al., 2000; Werner et al., 2000). While measured concentrations of chemical contaminants were generally below acutely toxic levels for fish, potential sublethal toxic effects resulting in, e.g. energy reallocation or behavioral abnormalities are of concern. Sublethal effects are difficult to detect in the field, but they may decrease the evolutionary fitness of fish populations (Scholz et al., 2000; Sorensen, 1991; De Vlaming et al., 2000; Sandahl et al., 2005).

Ecological effects of aquatic contaminants are difficult to detect and quantify. Available ecotoxicological tools for screening contaminant exposures in the field include bioassays, toxicity identification evaluations (TIEs), or risk assessments based on existing data (Rand, 1995; USEPA, 1989a,b, 1991, 2000). On a level of higher resolution, altered cellular and molecular responses to stressors can be used as powerful tools for gaining a better understanding of the mechanisms and toxicants involved, and thus as biomarkers for the identification of environmental impacts of contaminants on aquatic ecosystems (Huggett et al., 1992). The rising field of ecotoxicogenomics links the two disciplines: genomics and ecotoxicology, mostly by identifying cellular biomarkers and biosignals at the transcriptome level, thus creating sensitive indicators for the exposure to contaminants. In a first step, microarray approaches are used to initially identify suites of up- or down-regulated genes, and changes in gene expression of selected genes are quantified subsequently by quantitative real-time PCR. However, for non-model species the high number of unidentifiable genes from random libraries and the comparatively high costs of microarray development and use can pose substantial limitations to this approach. In addition, only few studies simultaneously consider multiple tissues and tissue-specific effects when carrying out studies on the transcriptome.

In the present study on striped bass (*M. saxatilis*) we chose a mechanistic approach, i.e. selecting and developing biomarkers for the detection of changes at the transcriptome level, based on well-defined biochemical toxicity pathways in fish and other vertebrates. In a first step, real-time TaqMan PCR biomarker systems were developed for a suite of three housekeeping genes (18S, L9, GAPDH) and seven stress response biomarkers for proteotoxicity (HSP70, HSP90), phase I detoxification mechanism (CYP1A1), metal-binding (metallothionein), immune-system functioning and pathogen-defense (TGF- β , Mx-protein, nRAMP). Striped bass are pelagic, euryhaline, apex predatory fish with Atlantic origin. Due to their popularity as sport fish, they are economically important and have been introduced to areas outside their natural distribution range such as the Sacramento-San Joaquin Delta and San-Francisco Bay of Cali-

fornia. However, abundance indices of age-0 striped bass in the Sacramento-San Joaquin Delta have shown dramatic declines in recent years (Stevens et al., 1985; Kimmerer et al., 2000, 2001).

The overall objective of this study was to assess the toxicity of two chemicals with different mechanisms of action, the heavy metal copper, and the pyrethroid insecticide esfenvalerate [(*S*)- α -cyano-3-phenoxybenzyl-(*S*)-2-(4-chlorophenyl)-3-methylbutyrate], at different levels of biological organization (mortality, growth, swimming behavior, and transcription levels of stress response genes) to striped bass. Copper is one of the most ubiquitous environmental pollutants worldwide, entering aquatic systems as a consequence of past and present mining activities, domestic effluents and runoff from agricultural and urban areas (Nriagu, 1979; Kayhanian et al., 2003). Copper is widely used as a fungicide in California, especially in vineyards, and has been found at high concentrations in environmental samples from the San-Francisco Bay area (De Vlaming et al., 2000, and references therein). Copper concentrations measured in northern California streams have ranged from 3.4 to 64.5 $\mu\text{g/L}$ (Eaton, 2007). The pyrethroid insecticide esfenvalerate has been shown to be present at toxic concentrations in agricultural stormwater runoff (up to 720 ng/L in orchard runoff; Werner et al., 2002a, 2004), and in sediments of agricultural and urban water bodies (Weston et al., 2004, 2005; Amweg et al., 2006). Esfenvalerate concentrations in Central Valley streams receiving winter storm runoff from fruit orchards ranged from trace level to 93 ng/L (Bacey et al., 2005).

We conducted this study using two reference toxicants with different mechanisms of action to compare the sensitivity of the commonly used mortality endpoint with sublethal responses such as altered swimming behavior and changes at the transcriptome level. We tested the hypotheses that copper and esfenvalerate have different effects on the transcription of selected stress response genes, and that various tissues (liver, spleen, white muscle, anterior kidney, and gills) show different quality and quantity of mRNA transcription.

2. Material and methods

2.1. Fish exposures

Juvenile striped bass were exposed to copper or the pyrethroid insecticide esfenvalerate [(*S*)- α -cyano-3-phenoxybenzyl-(*S*)-2-(4-chlorophenyl)-3-methylbutyrate, 98% purity, Chem Service Inc., West Chester, PA, USA] in two separate tests. Fish were exposed to $\text{CuCl}_2 \cdot \text{H}_2\text{O}$ (Sigma, St. Louis, MO, USA) for 7 days, and to esfenvalerate for 24 h, and mortality as well as sublethal endpoints (growth, swimming behavior, transcription of stress response genes) were quantified. The shorter exposure time for the esfenvalerate study is based on the hypothesis that this hydrophobic chemical tends to quickly adsorb to particulate and organic matter in a typical field situation (Yang et al., 2006a,b; Brady et al., 2006) thus rendering exposure times for fish relatively short.

Juvenile striped bass used in the Cu exposure (90 days old, fork lengths 5.0–5.4 cm) were purchased from Professional Aquaculture Services (Chico, CA). Slightly larger, but only

81-day-old offspring from the same broodstock (fork lengths 5.3–8.0 cm; provided by D.J. Ostrach, UC Davis) were used for the esfenvalerate exposure. Fish used in the copper exposure were slowly acclimated to experimental conditions (conductivity: $890 \pm 20 \mu\text{S}/\text{cm}$; hardness: $200 \text{ mg}/\text{L CaCO}_3$) over the course of 3 days before tests were initiated. The acclimation and control water was obtained from a local, approximately 60 m deep well, passed through a packed column aerator to remove excess nitrogen and re-oxygenate. Striped bass used in the esfenvalerate exposure were maintained in flow-through circular tanks containing well water treated as described above for 2 weeks before the tests. Previous fish exposures have shown that stress due to transport and maintenance in the laboratory following the procedures described above is minimal. Fish were loaded into experimental 2.5-gal aquaria 24 h prior to testing. Each experimental treatment was comprised of five replicate aquaria containing five animals each. Each tank contained 5 L of water at 20°C and was aerated throughout the experiment. Tests were initiated by replacing 80% of the water with experimental copper or esfenvalerate solutions, or control water to yield nominal concentrations of 0 (control), 50, 200, 500 and $1000 \mu\text{g}/\text{L Cu}^{2+}$, or 0 (control), $200 \mu\text{L}/\text{L MeOH}$ (solvent control), 1, 3, 7 and $10 \mu\text{g}/\text{L}$ esfenvalerate. Measured copper concentrations on day 0 were 42, 160, 470, and $900 \text{ ppb total Cu}^{2+}$, and 42, 160, 440, and $810 \text{ ppb dissolved Cu}^{2+}$. Measured esfenvalerate concentrations on day 0 were 0.64, 2.20, 4.40 and $6.50 \mu\text{g}/\text{L}$.

Experiments were conducted using a light:dark cycle of 16 h:8 h. During the 7-day copper exposure, fish were fed daily (Silver Cup 2.0 mm pellets). Approximately 80% of the water in each replicate was renewed on days 2, 4 and 6. On days 1, 3 and 5, the numbers of live, dead, and missing fish were scored for each replicate. For the 24 h esfenvalerate exposure, fish were not fed and no water exchange was carried out.

Water temperature (T), pH, ammonia nitrogen ($\text{NH}_3\text{-N}$), and dissolved oxygen (DO) were measured daily and at test termination. Measured mean values ($\pm\text{S.D.}$) were T : $20.4 (\pm 0.7)^\circ\text{C}$, pH: $8.2 (\pm 0.3)$, $\text{NH}_3\text{-N}$: $0.5 (\pm 0.4) \text{ mg}/\text{L}$, DO: $8.5 (\pm 0.4) \text{ mg}/\text{L}$ for the Cu exposure and T : $20.3 (\pm 0.4)^\circ\text{C}$, pH: $7.8 (\pm 0.3)$, $\text{NH}_3\text{-N}$: $0.3 (\pm 0.3) \text{ mg}/\text{L}$, DO: $8.2 (\pm 0.7) \text{ mg}/\text{L}$ for the esfenvalerate exposure. On days 2 and 3 of the Cu exposure, ammonia-N temporarily reached $0.8/1.1 \text{ mg}/\text{L}$ ($0.1/0.19 \text{ mg}/\text{L}$ unionized NH_3) in controls, $0.95/1.0 \text{ mg}/\text{L}$ ($0.15/0.14 \text{ mg}/\text{L}$ unionized NH_3) in the 50 ppb Cu treatment, and $1.37/1.07 \text{ mg}/\text{L}$ ($0.19/0.15$ unionized NH_3) in the 200 ppb Cu treatment. This was probably due to excess feeding during those days but did not cause mortality. Overall, no significant deviations between measured water parameters among treatments or replicates were detected.

The number of dead fish was counted at the end of the experiment and surviving fish were sacrificed using an overdose of the anesthetic MS-222 (Sigma, St. Louis, MO, USA) in ice water to minimize degradation of RNA. Fork length (to nearest mm) and weight (to nearest 0.1 g) of each fish were recorded. No significant differences in length or weight were detected between individual treatment groups and controls. During the esfenvalerate exposure, swimming behavior and mortality endpoints were assessed after 4 and 24 h. Swimming behavior was assessed by observing each tank for 5 min. Any pronounced deviation

(>1 min) from normal (control) swimming patterns was assessed to be abnormal, e.g. when fish were not able to maintain buoyancy, flipped to their sides, lay on the ground, or repeatedly swam in small circles.

2.2. Quantitative real-time PCR

2.2.1. Tissue preparation, RNA extraction and cDNA synthesis

Surviving individuals (control group, 42 ppb total Cu, 160 ppb treatments for Cu exposure; control, solvent control, 0.64 and $2.2 \mu\text{g}/\text{L}$ for esfenvalerate exposure) were sampled for subsequent analyses of sublethal biomarkers. Fifteen fish per treatment (three fish per replicate) were dissected immediately after individuals were sacrificed and measured. The entire gill apparatus, liver, spleen, anterior kidney and two pieces of epaxial muscle from the left flank (<30 mg) were removed, placed in sterile, RNase and DNase free 1.5 mL Eppendorf vials, and immediately snap-frozen in liquid nitrogen. Samples were stored at -80°C until RNA extraction and cDNA synthesis. Frozen tissue samples (approximately 10 mg of liver, muscle and gill, 9 mg total spleen and 4 mg total anterior kidney) were transferred to 1.5 mL collection tubes (RNeasy Mini Kit, Qiagen Inc., Valencia, CA), re-immersed in liquid nitrogen and ground to a fine powder with a sterile pestle. Subsequently, $350 \mu\text{L}$ of RNeasy lysis buffer (RLT, RNeasy Mini Kit, Qiagen Inc.) were added, and lysates were homogenized by pestle and by passing them through a pipette tip approximately 10 times. After incubation for 3 min at room temperature, the RNA was extracted according to the manufacturer's recommendations (RNeasy Mini Kit, Qiagen Inc.). Thereafter, $20 \mu\text{L}$ of each freshly extracted nucleic acid sample was digested with 10 U of RNase free DNase I (Roche, Mannheim, Germany) for 15 min at 37°C to remove genomic DNA. DNase digested RNA was quality controlled for absence of genomic DNA contamination. All samples had a minimal difference of 7 CT values between the cDNA and digested total RNA (tRNA), indicating that remaining gDNA contamination in the tRNA was 1% or less. Complementary DNA (cDNA) was synthesized using 100 units of SuperScript III (Invitrogen, Carlsbad, CA, USA), 600 ng random hexadeoxyribonucleotide (pd(N)6) primers (random hexamer primer), 10 U RNaseOut (RNase inhibitor), and 1 mM dNTPs (all Invitrogen, Carlsbad, CA, USA) in a final volume of $40 \mu\text{L}$. The reverse transcription reaction proceeded for 50 min at 50°C . After addition of $60 \mu\text{L}$ of water, the reaction was terminated by heating for 5 min to 95°C and cooling on ice.

2.3. Real-time TaqMan[®] PCR

A suite of new real-time TaqMan PCR systems for proteotoxicity (HSP70, HSP90), phase I detoxification mechanism (CYP1A1), metal-binding (metallothionein), immune-system functioning and pathogen-defense (TGF- β , Mx-protein, nRAMP) were designed for studying sublethal stress response at the transcriptome level (see Table 1). For each target gene, two primers and an internal, fluorescent-labeled TaqMan probe (5' end, reporter dye FAM (6-carboxyfluorescein), 3' end, quencher

Table 1
List of real-time TaqMan PCR systems developed for striped bass (*Morone saxatilis*)

Gene specification	Species for starting sequence	NCBI GenBank accession number	Primer sequences	Positions start–end	TaqMan probe number and sequence
Stress response genes					
HSP70	<i>Dicentrarchus labrax</i>	AY423555	F: CATCCTTCTGGGGACAAGTCAG R: ACACCTCCAGCGGTCTCAATAC	1251–1343	62, ACCTGCTG
HSP90	<i>Dicentrarchus labrax</i>	AY395632	F: GACGAATACTGTGTCCAGCAGTTG R: CAGCTCCAGACCCCTTTTGGT	1631–1708	54, CTGGTCTC
CYP1A1	<i>Dicentrarchus labrax</i>	AJ251913	F: GCGGCACAACCCAGAGTA R: CAGCTTTCATGACGGTGTGAG	461–556	65, CTGGAGGA
Metallothionein	<i>Morone saxatilis</i>	AF091100	F: GCGGAGGATCCTGCACTTG R: CAGCCAGAGGCACACTTGGT	41–140	68, CTGCTCCT
TGF- β	<i>Morone chrysops</i> \times <i>Morone saxatilis</i>	AF140363	F: ATGGTTAAGAAAAGCGCATTGAA R: TCCGGCTCAGGCTCTTTG	88–167	36, GGAGCCAG
Mx-protein	<i>Dicentrarchus labrax</i>	AY424962	F: GTTCATGGTCAAGGAGCAGATCA R: GCTGTATGAACACCTTTCTAACAGCAT	1254–1348	11, GCTGGAAG
nRAMP	<i>Morone saxatilis</i>	AY008746	F: CTACTTCATTGAGTCAACTATTGCTCTCTT R: TCATGCACTTCCATATTGGTTTTATT	867–977	12, CTCCTTCC
Housekeeping genes					
18S	<i>Dicentrarchus labrax</i>	AY831388	F: GGCCGCTTTGGTGACTCTAGATA R: GAAAGTTGATAGGCAGACATTTCG	142–230	149, AGGCGGCGA
L9	<i>Dicentrarchus labrax</i>	DT044997	F: AAGCTTCGTGTGGATAAATGGTG R: GCAGATGGTGGGACTGTG	166–228	12, HGGAAAGGAG
GAPDH	<i>Dicentrarchus labrax</i>	AJ567450	F: TGTCCACAGACTTCAACAGTGACC R: AAAGTGGTCGTTGAGAGCGATG	372–459	25, HCTCCTCCA

Gene specification, species and NCBI GenBank accession number based upon which systems were designed, primer sequences, amplicon start and end position, TaqMan probe number (Universal Probe Library, Roche), and probe sequence.

dye TAMRA (6-carboxytetramethylrhodamine)) were designed using Primer Express software (Applied Biosystems, Foster City, CA, USA). TaqMan-primer design was based on NCBI Gene Bank sequence information for *M. saxatilis* and related bass species. Three housekeeping gene TaqMan systems (L9, 18S, GAPDH) were designed, based on sequence information from *Dicentrarchus labrax*. All real-time TaqMan PCR systems were validated for specificity and amplification efficiencies as described in Leutenegger et al. (1999). Briefly, 2-fold dilution series of cDNA samples were tested in triplicate with the respective real-time TaqMan PCR system. From the slope of the standard curve, the amplification efficiency was calculated using the formula $E = 10^{1/s} - 1$. All amplification efficiencies were above 90%, validating the specificity of the marker systems. Real-time TaqMan PCR mixes contained 400 nM of each of two primers and 80 nM of the appropriate TaqMan probe. We used TaqMan Universal PCR Mastermix (Applied Biosystems, Foster City, CA, USA) containing 10 mM Tris-HCl (pH 8.3), 50 mM KCl, 5 mM MgCl₂, 2.5 mM deoxynucleotide triphosphates, 0.625 U AmpliTaq Gold DNA polymerase per reaction, 0.25 U AmpErase UNG per reaction and 5 μ L of the diluted cDNA sample in a final volume of 12 μ L. The samples were placed in 384 well plates and cDNA was amplified in an automated fluorometer (ABI PRISM 7900 Sequence Detection System, Applied Biosystems). Amplification conditions were 2 min at 50 °C, 10 min at 95 °C, 40 cycles of 15 s at 95 °C and

60 s at 60 °C. Fluorescence of samples was measured every 7 s and signals were considered positive if fluorescence intensity exceeded 10 times the standard deviation of the baseline fluorescence (threshold cycle, C_T). SDS 2.2.1 software (Applied Biosystems) was used to quantify transcription.

2.4. Relative quantification of stress response gene transcription

The comparative C_T method was applied to quantify gene transcription of investigated stress response genes (User Bulletin #2, Applied Biosystems). Values are reported as relative transcription or the n -fold difference relative to a calibrator cDNA (i.e. average target gene transcription of control fish). Three housekeeping genes (18S, L9, GAPDH) were tested and the one revealing smallest standard deviation and most stable transcription levels over all treatments (L9) was used to normalize the target gene signals (ΔC_T) for the differences in the amount of nucleic acid added to each reaction and the efficiency of the reverse transcriptase step. The ΔC_T for each experimental sample from the exposed fish was subtracted from the ΔC_T of the calibrator, the mean target gene signal of control fish. Finally, the linear amount of target molecules relative to the calibrator was calculated by $2^{-\Delta\Delta C_T}$. Therefore, all stress response gene transcriptions are expressed as an n -fold difference relative to the calibrator. For comparisons of basic linearized transcription

values between tissues of all pooled control fish, muscle tissue revealed lowest transcription levels in all stress response genes and average transcription of each stress response gene in muscle was thus used as a calibrator for other tissues.

2.5. Statistical analyses

Gene transcription data were first tested for normality and equality of variances. Since more than the randomly expected number of datasets was either not normally distributed or failed equality of variance tests, we generally used non-parametric methods for comparisons between treatments and tissues. Kruskal–Wallis one-way analysis of variance on ranks (K-W ANOVA) was used to detect differences in linearized mean responses between treatments and tissues. In case of significance ($p < 0.05$), we tested for (i) differences in gene transcription between control and treatment groups and (ii) differences in gene transcription between the tissue with the weakest transcription level and other tissues by using non-parametric Mann–Whitney U -test. For comparisons between tissue types, Bonferroni corrections were applied to adjust p -values for multiple comparisons.

We decided to use a conservative and non-parametric statistical approach throughout the dataset for simplicity and in order to reduce the number of false-positives. It should be noted, however, that the robustness of data interpretation is strengthened by the fact that these results were very similar to those obtained by using parametric tests (one-way analysis of variance, ANOVA and Dunn's or Tukey's post hoc tests) with the limitation that some comparisons could not have been carried out under the requirements for equality of variances and normal distribution. Statistical analyses were carried out using the statistical programs Statistica 6.0 (StatSoft Inc., Tulsa, OK, USA), SPSS 7.0 and SigmaStat 2.0 (SPSS, Inc., Chicago, IL).

Lethal and sublethal effective concentrations were calculated using CETIS v. 1.1.2 (Tidepool Scientific Software, McKinleyville, CA, USA, 2006). NOEC and LOEC were calculated using USEPA standard statistical protocols (USEPA, 2002). LC50s and EC50s were calculated using linear regression, non-linear regression, or linear interpolation methods. For each endpoint, toxicity is defined as a statistically significant difference ($p < 0.05$) to the laboratory control.

3. Results

3.1. Responses to Cu and esfenvalerate at the organism level

Summaries of the effect concentrations of Cu and esfenvalerate on survival of striped bass juveniles are provided in Tables 2 and 3. For the Cu exposure, 100% mortality was observed at 470 and 900 $\mu\text{g/L}$ Cu (440 and 810 $\mu\text{g/L}$ dissolved Cu), whereas all fish survived in control water and at 42 $\mu\text{g/L}$ Cu (40 $\mu\text{g/L}$ dissolved Cu). At 160 $\mu\text{g/L}$ Cu, survival was 92%. The NOEC and LOEC for 96 h and 7 days were the same. No significant effects of Cu exposure on growth or swimming behavior were observed. LC50 and EC25 for total and dissolved

Table 2

Effect concentrations of Cu^{2+} on striped bass (*Morone saxatilis*) survival during a 7-day exposure

Time	Total Cu^{2+} (ppb)			Dissolved Cu^{2+} (ppb)		
	LC50	NOEC	LOEC	EC25	NOEC	LOEC
96 h	441	160	470	414	160	440
7 days	262	160	470	254	160	440

Cu in the 7-day exposure were about 60% of those for the 96 h exposure.

Exposure to esfenvalerate for 24 h resulted in 100% mortality at 4.4 and 6.5 $\mu\text{g/L}$ esfenvalerate. At 2.2 $\mu\text{g/L}$ 24-h survival was 40%. All individuals of control, solvent control and 0.64 $\mu\text{g/L}$ esfenvalerate treatments survived, and only 1 out of the 25 solvent control fish (4%) showed abnormal swimming behavior. No mortality was observed after 4 h in any treatment, but abnormal swimming behavior was observed in 76% of striped bass exposed to 6.5 $\mu\text{g/L}$ esfenvalerate, and in 36% of fish exposed to 4.4 $\mu\text{g/L}$ esfenvalerate.

3.2. Responses to Cu and esfenvalerate at the molecular level

3.2.1. Housekeeping and stress response gene transcription across all tissues

Across all tissues and treatments, 18S showed highest transcription levels out of the three housekeeping genes tested in this study, followed by L9 and GAPDH. Pairwise comparisons between housekeeping genes revealed that transcription levels remained most constant for L9 in striped bass under various exposure conditions. Thus, all subsequent comparisons of stress response gene transcription are expressed as n -fold differences to the calibrator L9 gene. Transcription of sublethal stress markers HSP70, HSP90, CYP1A, TGF- β , MT, MX, and nRAMP was detected in all tissues (gills, liver, muscle, spleen and kidney tissue), but differed significantly between tissues in terms of both total transcription levels and the direction of the response (up-/down-regulation).

3.2.2. Tissue-specific effects of Cu

Significant changes in the transcription of stress response genes after 7-day Cu exposure were detected for four stress response genes in kidney tissue, for two genes each in spleen, muscle and gill tissue, and for one gene in liver tissue. A summary of transcription levels of stress response genes in investigated tissues of control, 42 and 160 ppb Cu exposed fish is

Table 3

Effect concentrations of esfenvalerate ($\mu\text{g/L}$) on striped bass (*Morone saxatilis*) survival and swimming behavior during a 24-h exposure

Time (h)	Survival			Swimming behavior		
	LC50	NOEC	LOEC	EC25	NOEC	LOEC
4	NA	6.5	>6.5	3.88	2.2	4.4
24	2.17	0.64	2.2	1.07	0.64	2.2

Table 4

Changes in mean tissue-specific stress response gene transcription levels and standard deviations (S.D.) of control, 42 ppb total Cu and 160 ppb Cu exposed *Morone saxatilis* expressed as *n*-fold linear differences to ribosomal L9 housekeeping gene transcription

Tissue	HSP70	HSP90	CYP1A1	MT	TGF- β	MX	nRAMP
Muscle							
Control mean	0.1	-0.2	-0.1	-0.1	-0.1	0.0	-0.2
S.D.	2.1	3.7	2.8	4.4	2.0	3.3	2.4
42 ppb Cu mean	-1.0	-1.2	-1.2	-0.8	-1.9*	0.5	-1.0
S.D.	1.9	1.4	2.5	1.4	3.0	3.7	2.3
160 ppb Cu mean	0.8	-1.0	-1.8	2.0*	-2.4**	-2.4	-0.4
S.D.	2.4	1.8	5.7	2.8	1.7	2.2	3.6
Spleen							
Control mean	-0.1	-0.1	0.2	0.2	0.1	0.0	0.1
S.D.	1.6	1.8	3.8	2.5	2.0	2.6	1.5
42 ppb Cu mean	0.8	-0.3	-1.2	3.7*	0.1	1.7	-0.8
S.D.	1.5	2.0	2.3	3.5	1.6	1.6	1.5
160 ppb Cu mean	0.7	0.6	-0.9	9.5***	1.9*	0.9	0.3
S.D.	1.6	2.1	2.1	7.0	1.4	2.4	1.5
Gill							
Control mean	0.0	0.0	0.1	-0.1	-0.2	0.2	-0.2
S.D.	2.0	1.9	2.4	5.3	3.6	2.0	1.6
42 ppb Cu mean	-0.5	-0.9	-1.6	6.4*	1.8	-1.0	0.4
S.D.	3.8	3.2	7.1	6.8	3.7	3.0	2.5
160 ppb Cu mean	-1.6	-1.1	0.1	2.4	1.4	-6.3***	-1.0
S.D.	3.5	4.3	8.5	3.9	5.5	5.9	4.1
Liver							
Control mean	0.1	0.1	0.0	-0.1	0.2	0.0	0.0
S.D.	3.2	3.4	5.0	3.1	3.5	4.1	4.0
42 ppb Cu mean	2.3	1.5	2.2	2.0	6.3**	3.2	0.9
S.D.	5.2	5.3	6.3	4.2	7.6	4.5	3.8
160 ppb Cu mean	2.3	3.4	0.4	2.5	4.0**	1.2	1.2
S.D.	3.1	3.7	2.8	5.3	2.5	3.7	2.7
Kidney							
Control mean	0.0	0.2	0.0	0.1	-0.2	0.0	0.3
S.D.	1.8	1.6	3.0	2.1	1.8	3.2	2.1
42 ppb Cu mean	1.9**	1.7	-0.2	1.0	0.5*	3.4	3.1*
S.D.	1.4	1.8	1.6	3.0	5.9	4.8	3.6
160 ppb Cu mean	1.8**	2.0	0.6	4.4***	2.7**	0.9	2.9**
S.D.	1.8	2.0	1.8	2.9	2.0	2.1	1.4

* ** and *** refer to significant differences of mean values at *p*-values of <0.05, <0.01 and <0.001, respectively; Mann–Whitney *U* post hoc tests were only carried out when Kruskal–Wallis ANOVA had *p* < 0.05.

provided in Table 4 and Fig. 1. Overall, the strongest changes in transcription levels were detected for metallothionein, which was on average (all tissues) up-regulated 2.5-fold following the 42 ppb Cu treatment, and 4.2-fold after the 160 ppb Cu treatment compared to control fish. The strongest effects on metallothionein transcription were observed in the spleen tissue, where transcription increased 3.7- and 9.5-fold in the 42 and 160 ppb Cu treatments, respectively. A positive correlation between Cu-concentrations in water and metallothionein expression was found in all tissues except gills. However, in the high-surface gill tissue, being most directly and intensely exposed to dissolved copper, metallothionein transcription had its peak (6.4-fold increase) at 42 ppb Cu, and transcription levels were lower in the 160 ppb treatment group (2.4-fold increase).

Significant changes in gene transcription levels were also detected for TGF- β , indicating a link between heavy metal exposure and the immune response. TGF- β gene transcription showed significant increases in liver and kidney tissue after

exposure to 42 and 160 ppb Cu, and following 160 ppb Cu in spleen tissue. In contrast, TGF- β transcription was significantly reduced in muscle tissue (-1.9 at 42 ppb and -2.4 at 160 ppb). In gill tissue, TGF- β transcription remained constant, but transcription of the cytokine MX was significantly decreased after exposure to 160 ppb Cu. The transcription of nRAMP remained constant in most tissues, but was significantly increased by a factor of 3 in kidney following both sublethal copper treatments. Although significant changes in the average transcription levels of heat shock proteins between treatments were only detected for HSP70 in kidney tissue, strongest individual effects for HSP70 and HSP90 occurred in liver tissue. As expected, the transcription of the phase-1 detoxification enzyme CYP1A1 was not affected by any tested Cu concentration in any tissue.

3.2.3. Tissue-specific effects of esfenvalerate

Overall, exposure of striped bass to esfenvalerate for 24 h resulted in less pronounced changes in gene transcription than

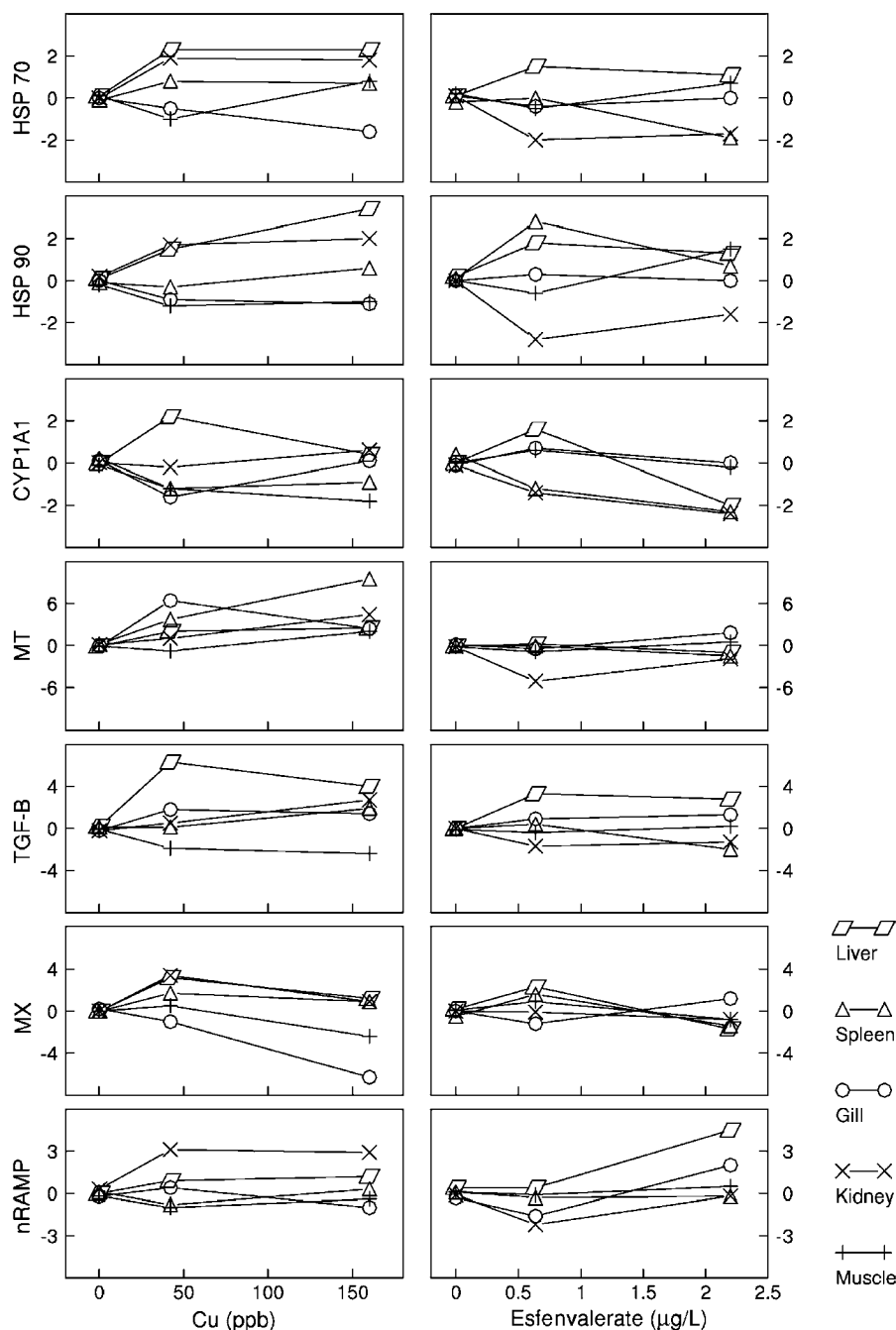


Fig. 1. Graphic overview showing changes in mean tissue-specific stress response gene transcription levels for Cu and esfenvalerate exposed *Morone saxatilis*, expressed as *n*-fold linear differences to ribosomal L9 housekeeping gene transcription; for details and standard deviations see Tables 4 and 5.

those observed after Cu treatments. A summary of changes in gene transcription after esfenvalerate exposure is given in Table 5 and Fig. 1. Significant alteration of transcription was detected in spleen and liver. Only two of the genes investigated were affected in kidney, and no changes were found in muscle and gill tissue. Most pronounced effects were detected for HSP90, with a 2.8-fold increase of transcription in spleen tissue. Transcription of HSP70 was up-regulated in the liver but down-regulated in spleen tissue. The same pattern was evident for CYP1A1. TGF- β transcription was increased 3.3-fold on average in liver tissue, corroborating our finding that it also showed strongest transcription increases in liver tissue following Cu exposure. Overall,

strongest effects of esfenvalerate exposure on gene transcription were observed at 0.64 $\mu\text{g/L}$, whereas weaker and insignificant changes of transcription were seen at 2.2 $\mu\text{g/L}$. Fish exposed to methanol (MeOH) (solvent control) and control fish had similar transcription values and showed no significant differences of transcription levels in any of the tissues investigated.

3.2.4. Comparison of gene transcription levels between tissues

A moderate to very high variation of gene transcription for individual stress response genes was observed between tissues (Fig. 2). Baseline levels of stress gene transcription for pooled

Table 5

Changes in mean tissue-specific stress response gene transcription levels and standard deviations (S.D.) of control, 0.64 and 2.20 $\mu\text{g/L}$ esfenvalerate exposed *Morone saxatilis* expressed as *n*-fold linear differences to ribosomal L9 housekeeping gene transcription

Tissue	HSP70	HSP90	CYP1A1	MT	TGF- β	MX	nRAMP
Muscle							
Solv. control mean	0.2	0.0	0.0	-0.2	-0.1	0.1	0.1
S.D.	2.4	2.9	3.4	4.0	2.3	2.4	4.2
0.64 $\mu\text{g/L}$ esfenv. mean	-0.5	-0.6	0.6	-0.9	-0.4	0.9	-0.1
S.D.	1.9	2.6	3.5	3.4	2.2	4.0	2.2
2.20 $\mu\text{g/L}$ esfenv. mean	0.7	1.5	-0.2	0.5	0.2	-0.8	0.5
S.D.	1.8	3.0	1.9	4.8	2.2	2.5	3.2
Spleen							
Solv. control mean	-0.2	0.1	0.4	0.0	0.0	-0.5	0.1
S.D.	1.8	2.5	5.1	2.0	2.1	2.2	2.0
0.64 $\mu\text{g/L}$ esfenv. mean	0.0	2.8**	-1.2	-0.1	0.4	1.6*	-0.3
S.D.	1.6	1.6	2.9	2.6	1.9	2.2	2.2
2.20 $\mu\text{g/L}$ esfenv. mean	-1.9**	0.7	-2.3*	-1.5	-2.0	-1.4	-0.2
S.D.	0.6	1.9	3.0	1.4	1.0	1.8	1.8
Gill							
Solv. control mean	0.1	0.0	-0.1	0.1	0.0	0.0	-0.3
S.D.	1.9	2.8	2.1	1.6	2.2	2.7	2.4
0.64 $\mu\text{g/L}$ esfenv. mean	-0.4	0.3	0.7	-0.5	0.9	-1.2	-1.6
S.D.	3.5	3.9	4.1	6.1	3.4	3.7	4.0
2.20 $\mu\text{g/L}$ esfenv. mean	0.0	0.0	0.0	1.8	1.3	1.2	2.0
S.D.	2.3	3.7	3.7	2.9	2.1	2.4	2.4
Liver							
Solv. control mean	0.1	0.2	0.0	-0.1	0.0	0.2	0.4
S.D.	1.3	2.0	1.7	2.3	3.7	2.7	4.5
0.64 $\mu\text{g/L}$ esfenv. mean	1.5**	1.8*	1.6*	0.2	3.3**	2.3	0.4
S.D.	0.9	0.3	1.5	1.7	1.5	5.2	2.9
2.20 $\mu\text{g/L}$ esfenv. mean	1.1	1.3	-2.0	-1.0	2.8	-1.7	4.5
S.D.	1.9	2.0	5.4	3.1	1.6	3.5	5.7
Kidney							
Solv. control mean	0.1	0.0	-0.1	-0.2	0.0	0.0	-0.1
S.D.	3.3	2.9	2.8	3.3	2.8	2.9	2.6
0.64 $\mu\text{g/L}$ esfenv. mean	-2.0	-2.8	-1.4	-5.1*	-1.7	-0.1	-2.2*
S.D.	1.6	1.4	2.0	5.2	1.8	3.4	2.8
2.20 $\mu\text{g/L}$ esfenv. mean	-1.7	-1.6	-2.4	-1.9	-1.3	-0.8	-0.2
S.D.	2.8	2.8	2.0	2.8	3.4	4.7	2.0

* ** and *** refer to significant differences of mean values at *p*-values of <0.05, <0.01 and <0.001, respectively; Mann–Whitney *U* tests post hoc tests were only carried out when Kruskal–Wallis ANOVA had *p* < 0.05.

control and solvent control fish were lowest in muscle tissue, thus average transcription for respective stress response genes in this tissue was used as a calibrator reference for comparisons with other tissues. Transcription of HSP70 and HSP90 was on an average 5–10-fold enhanced in spleen, gill, and kidney in comparison to muscle tissue. CYP1A1 showed most pronounced tissue-specific effects with up to 1000-fold stronger transcription in liver than in muscle. Metallothionein transcription was highest in liver, kidney and spleen tissues, whereas cytokine (TGF- β , MX, nRAMP) transcription was most pronounced in spleen.

4. Discussion

This study provides new information on the lethal and sub-lethal effects of two prominent water contaminants, the heavy metal copper and the pyrethroid insecticide esfenvalerate, on juvenile striped bass, a non-model fish species for which little is known about its sensitivity to pollutants. By quantifying

effects across different levels of biological organization, we provide information on the links between molecular responses and ecologically relevant effects.

The contaminants used in this study, copper and esfenvalerate, are known to be toxic to fish, but have very different mechanisms of action. Copper, an abundant heavy metal in the environment (Bielmyer et al., 2006), exerts its toxicity to fish by inhibiting the branchial Na^+K^+ -ATPase and ion uptake as well as stimulating Na^+ , K^+ and Cl^- efflux from gill surfaces (Lauren and McDonald, 1985). The pyrethroid insecticide esfenvalerate is a potent neurotoxicant that interferes with nerve cell function by interacting with voltage-dependent sodium channels as well as other ion channels, resulting in repetitive firing of neurons and eventually causing paralysis (Bradbury and Coats, 1989). It has been previously shown that juvenile hybrid striped bass (*Morone chrysops* \times *M. saxatilis*) are relatively sensitive to Cu exposure if acclimated to freshwater, with 96-h acute median lethal concentrations of 94 $\mu\text{g/L}$ (Bielmyer et al., 2006). This concentration

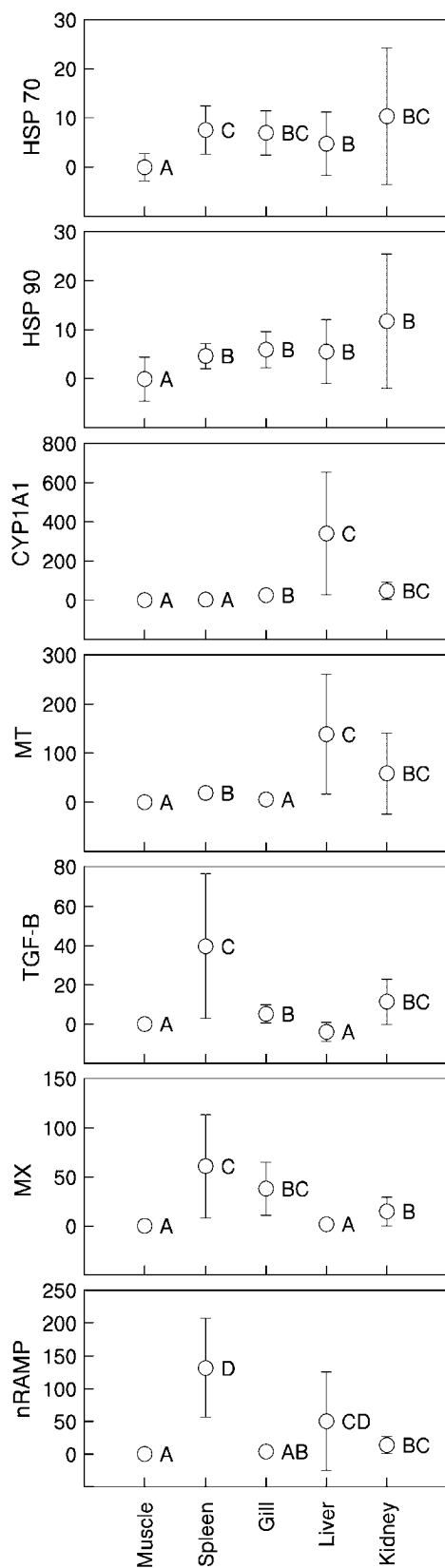


Fig. 2. Comparison of mean gene transcription profiles between tissues and standard deviation (S.D.) for stress response genes of all pooled striped bass (*Morone saxatilis*) from control treatments using muscle tissue from control fish as an overall calibration reference. Different letters indicate significant differences of $p < 0.05$.

is lower than the values observed in this study (414 $\mu\text{g/L}$), but toxicity of Cu in fish is strongly influenced by environmental parameters such as DOC, pH, hardness and salinity (Reardon and Harrell, 1990; Welsh et al., 1995; Erickson et al., 1996). Acute toxicity of esfenvalerate in fish occurs at concentrations of approximately 0.1–0.5 $\mu\text{g/L}$ (24–96-h LC50; Siepmann and Holm, 2000; Oros and Werner, 2005). Due to the compound's hydrophobic properties, exposure of aquatic organisms living in the water-column may only be brief (a few hours) or take place via dietary uptake (Werner et al., 2002b). The available data suggests that esfenvalerate toxicity to fish is size-dependent. This may explain why the 24-h LC50 of 2.17 $\mu\text{g/L}$ for striped bass juveniles used in this study was higher than reported values.

Sublethal toxic effects can occur at exposure levels far below the concentrations that cause lethality and can have severe consequences for the fitness, reproductive success and survival of aquatic organisms, ultimately leading to population-level effects (Carson, 1962). For an assessment of the toxic potential of chemicals on fish and aquatic ecosystems, endpoints from laboratory tests such as growth, swimming behavior and molecular stress responses should therefore be interpreted in the context of their environmental consequences. As confirmed in this study, growth endpoints are of limited value for short-term (<7 days) exposures of juvenile striped bass, especially if fish size is not homogenous and if the number of organisms tested must be limited to low numbers. Monitoring of swimming behavior can be a powerful and sensitive biomarker for sublethal effects, as shown for the esfenvalerate exposure. Decreased swimming performance most likely decreases the ability to chase prey or to avoid predation, and is thus an important indicator for overall fitness (Holcombe et al., 1982; Little et al., 1990; Scholz et al., 2000; Sandahl et al., 2005). Non-technical and non-computational methods for the assessment of abnormal swimming behavior, however, are prone to a certain bias depending on the researcher and the time intervals in which they are carried out, and are thus difficult to standardize. Linking results from laboratory exposures to field data is complicated by the fact that it mostly remains untested if fish are able to sense certain chemicals and minimize their exposure by swimming into refuge areas or if they become more vulnerable to predation.

Molecular endpoints are the most sensitive biomarker tools to identify and discriminate between exposures to groups of chemicals with different mechanisms of action, detoxification pathways, and biochemical stress responses. Our results demonstrate that analysis of even a limited set of molecular markers can be a useful approach in the field of "environmental forensics". The low amount of tissue needed for analysis (in the order of 2–10 mg), the short time lag between exposure and response, and the cost-effectiveness of the method are additional advantages. Toxicant-specific markers (e.g. metallothionein as an indicator for heavy metal exposure) may be most useful for toxicant identification, whereas integral pleiotropic markers that interact with different systems in diverse ways (e.g. HSP, see Basu et al., 2002) are most powerful for a general screening of overall stress in fish. It has to be noted, however, that for many model organisms and for all non-model organisms, the consequences of the complex temporal patterns of up-/down-regulation of mRNA

transcription and their link with proteomics and organismic effects are still poorly understood, particularly for long-term and repeated exposures. In addition, environmental stressors may induce responses and effects at different levels of organization. Thus, linking molecular responses with ecologically relevant parameters remains challenging.

Under environmental conditions it is likely that multiple stressors simultaneously impact fish populations, and that mixture effects between different chemical contaminants, physical stressors and pathogens may occur and exacerbate overall deleterious effects (Eder et al., 2007). Unknown “side effects” of pollutants, such as alteration of the immune response as observed in this study, behavioral changes, or interference of contaminants with the reproductive system may greatly reduce an organism’s fitness in the wild. For instance, synergistic effects between the pyrethroid insecticide esfenvalerate and viral susceptibility have been described in fish (Eder et al., 2007; Clifford et al., 2005). Also, indirect contaminant-induced effects, such as changes in trophic cascades (Fleeger et al., 2003) may influence populations and communities in nature and can only be detected if the most sensitive species are considered in monitoring.

The bioindicative function of transcriptomics is complicated by the high variability of direction and intensity of changes in gene transcription found in this study. Age and size of the organisms studied can strongly influence sensitivity, which may partly explain the highly variable transcriptome responses. Generally, one of the main limitations in studying “natural” populations is most likely the considerable degree of variation in gene transcription and expression levels between individuals and populations, which was evident for striped bass in this study, and which has been described before for other natural fish populations (e.g. Oleksiak et al., 2002). It is likely that variation in gene expression has a strong impact on evolution (King and Wilson, 1975) and that natural populations have the ability to adapt to certain levels of environmental stressors.

It is also essential that tissue-specific effects be taken into account when assessing an organism’s overall response to environmental stressors. In fact, there are usually various cell types within tissues, and a mere shift in the relative percentage of these can influence the intensity of the response. For instance, copper exposure for 7 days was described to cause a decrease in the number of kidney leukocytes in zebrafish (Rougier et al., 1994), suggesting that dilution or concentration effects can potentially disguise cellular response patterns if whole organs are analyzed. For routine applications and field screenings, however, investigations beyond the tissue level are at present not realistic and were thus not considered in this study. From a biomarker perspective, focusing on relative changes in gene transcription of stress response genes is most relevant, whereas an organism’s overall response will most likely better match the ecological relevance.

Both enhancement and silencing of gene transcription of stress response genes provide important mechanistic information on the organismal effects of contaminants. For example, the observed pronounced effects of Cu exposure on transcription of the cytokine TGF- β in several tissues suggest that there is a link between Cu and an altered immune response. This

result confirms earlier studies, where a link between exposure to Cu and cellular and non-specific immune-function was reported (Zelikoff, 1993; Rougier et al., 1994; Dethloff and Bailey, 1998; Dethloff et al., 2001; Shariff et al., 2001; Broeg, 2003). Furthermore, transcription of genes associated with the immune system was down-regulated in *Daphnia* sp. following Cu exposure (Poynton et al., 2007). Exposures to environmental stressors that have the potential of modulating the immune system can often be linked to ecologically relevant endpoints, such as decreased resistance to disease (Peters and Schwarzer, 1985; Anderson, 1990; Rougier et al., 1994; Dunier, 1996; Pulsford et al., 1995; Dethloff and Bailey, 1998; Dethloff et al., 2001). Corroborating existing information on the effects of Cu exposure on protein levels of MT (Hamer, 1986; Lauren and McDonald, 1987), transcription of this metal-binding protein was significantly induced in response to Cu treatment. However, induction was more pronounced in hematopoietic tissues (spleen, kidney) than in liver, where protein levels are commonly measured. Our data underline the potential of using MT induction on the transcriptome level as a biomarker, especially since the availability of sufficient amounts of tissue can quickly become a challenge when quantifying proteins in spleen or kidney of small fish. In addition, signals at the transcriptome level generally take less time to manifest themselves. Combining studies into the transcriptome as well as proteome of stress response mechanisms in fish can thus be useful for separating and assessing the spheres of effect and response. This study also shows that higher concentrations of Cu and esfenvalerate can result in weaker responses on the transcriptome level than lower concentrations, indicating that receptor saturation (high dose inhibition) or cellular damage may play a role, leading to potentially irreversible effects. For heavy metals, stimulation of macrophages at lower exposure concentrations and inhibition at high concentrations has been previously reported (e.g., Pulsford et al., 1995).

Our findings for the non-model organism striped bass suggest that mechanistic approaches in transcription biomarker studies, i.e. hypothesis-driven approaches of investigating gene transcription in pre-selected target genes for a large number of individuals in various tissues, can be an effective alternative to microarray studies, where costs can severely limit the number of tissues and individuals to be used, and where interpretations of up- or down-regulation often cannot be linked to known biochemical pathways. Molecular endpoints, i.e. the change in transcription levels of stress response genes, were more sensitive indicators for exposure to model contaminants than other sublethal (growth, swimming behavior) or lethal (mortality) endpoints. These results indicate that changes in transcription of selected stress response genes can be powerful biomarkers for the detection of toxic effects at sublethal levels, and for the identification of pollutants. Our findings demonstrate that it is possible to carry out functional genomic experiments in the context of ecotoxicogenomics, even in species with limited availability of sequence information. The strong dependence of the observed transcriptome response on tissue type and dose, and the fact that organisms are often exposed to multiple stressors simultaneously, still pose challenges to the routine use of this technique for field sample assessment.

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