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Pelagic Organism Decline (POD): Acute and Chronic Invertebrate and Fish Toxicity Testing in the Sacramento-San Joaquin Delta 2008-2010

Final Report

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I. Executive Summary

The study described here encompasses (1) a 2-year toxicity monitoring program in the Sacramento-San Joaquin (SSJ) Delta, including several sites in Suisun Bay and the Napa River; (2) laboratory investigations on the comparative sensitivity of important fish and aquatic invertebrate species to chemical contaminants of concern in the Delta; and (3) studies to develop biomarker tools for fish species of special interest.

1. Toxicity Monitoring

From January 1, 2008 to December 31, 2009, the UC Davis Aquatic Toxicology Laboratory and California Department of Fish and Game collected (biweekly) 752 water samples from 16 sites in the Northern Sacramento-San Joaquin Estuary. Toxicity tests were performed using *Hyalella azteca,* an amphipod species resident in the Delta, with survival and growth as acute and chronic endpoints, respectively. Routine partial toxicity identification evaluation (TIE) tests were conducted on all water samples with piperonyl-butoxide (PBO), a chemical synergist/antagonist, to provide early evidence for the presence of two classes of toxic insecticides: pyrethroids and organophosphates. During spring (March – May), water samples from five sites (340, Cache-Lindsay, Hood, Light 55, and Suisun) were tested for toxicity to larval delta smelt (*Hypomesus transpacificus*). Exposure systems for *in situ* monitoring (continuous flow toxicity testing on-location at field sites) were designed and tested at two DWR water quality monitoring stations (Rough & Ready Island - San Joaquin River; Hood - Sacramento River). Test organisms were *H. transpacificus*, fathead minnows (*Pimephales promelas*), and *H. azteca*. Laboratory test methods were developed for the calanoid copepod, *Eurytemora affinis*.

Water Quality at Field Sites: Water quality parameters were routinely monitored in ambient water samples. Field water temperature ranged from 6.8 (Cache-Ulatis) to 29.8^{o}C (Rough & Ready Island), dissolved oxygen from 5.1 (Suisun Slough at Rush Ranch) to 14.9 mg/L (site 711 – Rio Vista), electrical conductivity from 101 (site 711) to 24,965 µS/cm (Napa River), pH from 6.43 (site 405 - Carquinez Strait) to 8.70 (Napa River) and turbidity from 2 (multiple sites) to 713 NTU (Napa River). Highest total ammonia/um concentrations were 0.62 mg/L at site 405; 0.59 mg/L at Hood and 0.58 mg/L at site 711 (Rio Vista). Un-ionized ammonia concentrations were highest at stations Cache-Lindsey and 711 with maximum concentrations of 0.025 and 0.021 mg/L, respectively. During the project period, water at Hood, 711 and Cache-Lindsey had significantly higher total ammonia/um and un-ionized ammonia concentrations than all other study sites, overall.

Toxicity Monitoring with H. azteca: Four water samples (0.5% of total) from sites 711, Light 55, 405 and Suisun Slough at Rush Ranch, were acutely toxic to the amphipods. Two of these (site 711 and Suisun Slough) had less than 50% survival. Addition of PBO increased acute toxicity in seven ambient samples (0.9% of total) suggesting the presence of pyrethroid insecticides. Three of these contained detectable concentrations of pyrethroids: cypermethrin (site 902 – Old River, Aug. 28, 2008); bifenthrin and lambda-cyhalothrin (Cache-Ulatis, Feb. 28, 2008); and permethrin (Hood, Jun. 23, 2009). Although toxicity identification evaluation indicated that pyrethroid insecticides were the dominant toxicants in other samples from Cache-Ulatis (collected Jan. 31, 2008 and Oct. 15, 2009), and from Suisun Slough (collected on August 12,

2008), pyrethroids were below detection limits. We conclude that loss of pyrethroid pesticides due to degradation and adsorption poses problems for the confirmation of toxicity testing results by analytical chemistry at concentrations that can cause toxic effects in sensitive invertebrates (for details see Chapter IV.4).

Chronic toxicity in the form of reduced amphipod growth compared to controls was detected in 7 samples (0.9 % of total) without apparent seasonal or geographic patterns. Amphipods exposed to a sample from Cache-Ulatis (collected Jan. 31, 2008) not only grew significantly less than control animals, but PBO addition also synergized acute toxicity of the ambient sample (see above). This shows that effects seen after PBO addition ("PBO effects") can signal the presence of pyrethroid insecticides at concentrations that cause sublethal toxicity. A total of 100 samples (13.3 %) showed significant PBO effects on amphipod growth, 45 in 2008 and 55 in 2009. Of these, 36 (4.8% of total samples tested) showed an increase in weight with PBO addition ("antagonistic PBO effect"), and 64 (8.5% of total) a decrease ("synergistic PBO effect"). Overall, Cache-Ulatis had the greatest number of samples demonstrating PBO effects on survival suggested the presence of pyrethroid insecticides. Sites Hood, Cache-Ulatis, Rough and Ready Island, Suisun Slough at Rush Ranch, Light 55, 915 and 508 had the greatest number of samples with PBO effects on amphipod growth.

A total of 113 water samples were submitted for chemical analysis. Pyrethroids, in particular cyfluthrin (≤20 ng/L), permethrin (≤35 ng/L), cypermethrin (≤16 ng/L), bifenthrin (≤117 ng/L) and esfenvalerate (≤9 ng/L), were detected in 24 samples, organophosphates, in particular chlorpyrifos (≤10 ng/L), diazinon (≤12 ng/L) and disulfoton (≤17 ng/L), were detected in 13 samples, and the herbicide diuron (≤86 ng/L) was detected in all ten samples it was analyzed in. Relatively high concentrations of dissolved copper (4.4-4.9 µg/L) were detected in water samples from Cache-Lindsey and Rough and Ready Island, both freshwater sites. Numerous samples contained more than one contaminant.

Effect of ammonia/um on amphipod survival and growth: Correlation analysis of data collected over a 4-year monitoring period (2006-2010) revealed significant relationships between amphipod survival and growth at several sites. Total ammonia/um concentrations were negatively correlated with amphipod survival at Cache-Ulatis. Amphipod growth negatively correlated with total ammonia/um at sites 405, 609, 711, Light 55, Napa and Rough and Ready Island, and with un-ionized ammonia at Rough and Ready Island.

Toxicity Monitoring with larval delta smelt: Results of tests conducted in March – May, 2008, suggest that among six sampling sites (340 - Napa River, Cache-Lindsey, Hood, Light 55, and Rough & Ready Island), water quality at Hood was least favorable for larval delta smelt. Water from Hood generally had the lowest EC and turbidity. Even though control treatments included water adjusted to the lowest EC or low turbidity, these two parameters combined may have caused or contributed to the high mortality observed in water samples from Hood. Overall, survival was highest in water from Suisun Slough, the Napa River and Light 55.

In 2009, antibiotics were added to all tests due to concerns regarding disease compromising control survival. This measure improved delta smelt control survival in

2009 tests. Additional control treatments were added to account for turbidity and/or EC effects, which showed that the combination of these two stress factors was more stressful to fish than each factor by itself. Survival was significantly lower than controls in three samples collected from the San Joaquin River at Rough & Ready Island. This was attributed to toxic contaminants in the sample collected on May 14, 2009, however, low turbidity as a contributing stress factor cannot be ruled out in the other two. In addition, samples from Hood and Cache Lindsey on April 29, 2009, were acutely toxic to larval delta smelt. As in 2008, water from Suisun Slough had the highest survival, overall.

We conclude that water quality in the Sacramento River at Hood, in the San Joaquin River at Rough & Ready Island, and near the confluence of Cache and Lindsey Sloughs is at times unfavorable for larval delta smelt. This may in some cases be partly attributable to low turbidity stress. Turbidity, and to a lesser degree salinity/EC, are extremely important parameters influencing larval delta smelt survival. All testing therefore requires controls matching the salinity/EC and turbidity of each sample to obtain conclusive data on water toxicity.

In Situ Monitoring: A sophisticated exposure system was designed and installed for simultaneous *in situ* exposures of multiple fish and invertebrate species at two DWR water quality monitoring stations located in Hood, (Sacramento River) and Rough & Ready Island in Stockton (San Joaquin River). During March - May, 2009, six exposure experiments were conducted using larval delta smelt, larval fathead minnow, and the amphipod *H. azteca*, concurrently with ambient delta smelt toxicity testing at UCD-ATL. Larval *H. transpacificus* survival was poor both in the control and ambient water. No toxicity to *H. azteca* or *P. promelas* was detected.

Toxicity Monitoring with Copepods: One 7-d toxicity pilot test using juvenile *E. affinis* was conducted in May 2009 with samples from sites 711, Cache-Ulatis, Hood, and Light 55, and a series of low EC controls (100, 250, 500, 1000, 1900 µS/cm). Results showed that EC can significantly affect copepod survival. Survival was highest in the 1900 µS/cm control and decreased with decreasing conductivity. Survival was low in ambient samples with the exception of Cache-Ulatis (100% survival) possibly due in part to the available food and/or higher turbidity.

2. Comparative Species Sensitivity

Toxic effect concentrations for larval delta smelt, larval fathead minnow, the amphipod *H. azteca*, the waterflea *Ceriodaphia dubia,* and the copepod *E. affinis* were determined for a series of chemical contaminants of interest in the Sacramento-San Joaquin Delta, including organophosphate (chlorpyrifos, diazinon) and pyrethroid (bifenthrin, cyfluthrin, esfenvalerate, permethrin) insecticides, copper, and ammonia.

Larval delta smelt were 1.8 to >11-fold more sensitive than fathead minnow to ammonia/um, copper, and all insecticides tested with the exception of permethrin.

E. affinis and *C. dubia* were most sensitive to ammonia/um, and *C. dubia* was most sensitive to copper. Waterfleas were the most sensitive, and copepods the least sensitive species with respect to the organophosphates, chlorpyrifos and diazinon. Amphipods were the most sensitive to all pyrethroid insecticides tested.

Among toxicity test endpoints and species, *C. dubia* reproduction was the most sensitive with regard to copper and organophosphate insecticides, while *H. azteca* 10-d survival and growth were the most sensitive for detecting pyrethroid toxicity. The *C. dubia* chronic endpoint (reproduction) was far less sensitive than the acute endpoint (96-h survival) for pyrethroid toxicity, likely due to the differences in test protocols. An in-depth analysis of test methods is provided by Deanovic et al. (Chapter IV.4).

3. Sublethal Indicators of Contaminant Effects

Molecular biomarkers were developed to examine changes in gene expression after exposure to ambient water or known sublethal concentrations of contaminants. A cDNA microarray for delta smelt was developed with 8,448 Expressed Sequence Tags (ESTs) to study the effects of individual chemical stressors and exposure to Delta water samples; and to select mechanism-based biomarker genes for inclusion in mRNA-arrays. Biomarker responses were linked to ecologically relevant physiological effects. To date, gene responses were measured in larval and juvenile fish exposed to the pyrethroid insecticide esfenvalerate, copper and ammonia/um. Results from these studies support the use of functionally characterized suites of molecular biomarkers (mRNA arrays) to assess the effects of contaminants on aquatic species.

Biomarker Development for Delta Smelt - Esfenvalerate: Exposure to esfenvalerate affected swimming behavior of larval delta smelt at concentrations as low as 0.063 $\mu g.L^{-1}$, and significant differences were seen in expression in genes involved in neuromuscular activity (Connon et al. 2009; Chapter V.1). Genes associated with immune responses, apoptosis, redox, osmotic stress, detoxification, and growth and development were also affected. Swimming impairment correlated significantly with expression of aspartoacylase (ASPA), an enzyme involved in brain cell function and associated with numerous neurological diseases in humans. Selected genes were investigated for their use as molecular biomarkers, and strong links were determined between measured downregulation in ASPA and observed behavioral responses in fish exposed to environmentally relevant pyrethroid concentrations.

Biomarker Development for Delta Smelt - Copper: Differences in sensitivity to copper were measured between juvenile and larval delta smelt ($LC_{50\text{-}96h}$= 25.2 and 80.4 $\mu g.Cu^{2+}.L^{-1}$ respectively; Connon et al., Chapter V.2). Swimming velocity declined on exposure in a dose dependent manner. Genes encoding for ASPA, hemopexin, alpha-actin and calcium regulation proteins were significantly affected by exposure and were functionally linked with measured swimming responses. Effects on digestion were measured by upregulation of chitinase and downregulation of amylase. Downregulation of tumor necrosis factor indicated a compromised immune system.

Biomarker Development for Delta Smelt - Ammonia/um: A four-day exposure of 57-day old larvae resulted in a measured un-ionized ammonia (NH_3) LC_{50} of 147$\mu g.L^{-1}$, a NOEC of 66 $\mu g.L^{-1}$ and LOEC 105 $\mu g.L^{-1}$. Genome-wide gene expression was measured at 105 $\mu g.L^{-1}$ and selected genes of interest were further investigated using quantitative PCR analyses. Genes predominantly encoding for membrane bound proteins responded significantly to ammonia exposure, however, neurological and muscular activity were also impaired (Connon et al., Chapter V.3).

Biomarker Development for Striped Bass: To develop biomarker tools for striped bass (*Morone saxatilis*), the dynamics of CYP1A induction at different biological levels (gene expression / protein level / catalytic activity) were investigated (Connon et al., Chapter V.4). Specific objectives were to define the respective ranges of utilization as biomarkers of PAH contamination in this species, and to better understand the relationship of CYP1A gene expression with higher level cellular effects. Cytochrome P4501A (CYP1A) is a monooxygenase involved in xenobiotic biotransformation and detoxification.

Results showed that CYP1A gene expression responded rapidly to PAH exposure, and corresponding biotransformation and detoxification proteins were synthesized within the first few hours of exposure. CYP1A mRNA transcription was significantly upregulated in a dose response manner following 24 h post BNF injection, reaching a plateau at 25 $mg.kg^{-1}$ at 180-fold change induction. Significant differences were measured at concentrations as low as 0.1 $mg.kg^{-1}$, resulting in a 12-fold change in gene expression. This study demonstrated that methods used to measure the effect of PAH exposure at the molecular, protein and catalytic activity levels were successful in detecting CYP1A induction in striped bass. Models utilizing the differences in responses measured by each of the three biomarkers could be used to detect, in field situations, signatures of past and/or current exposure.

In addition, tissue samples from juvenile striped bass exposed to extracts of SPMD membranes deployed at several sites in the SSJ Delta were analyzed for expression of four stress-responsive genes, vitellogenin, CYP1A, metallothionein and hsp70 (Connon et al., Chapter V.5). Results provide preliminary indication of general stress and immune system responses at Boynton Slough and Sherman Lake sites.

Linking Molecular Biomarkers to Higher Level Effects in Fathead Minnow: Beggel et al. (2010; Chapter V.6) describe a study on the lethal and sublethal toxicity of commercial pesticide formulations and their active ingredients to larval fathead minnow (*P. promelas*). This study compared toxicity of two current-use insecticides, the pyrethroid bifenthrin, and the phenylpyrazole fipronil, to their commercial formulations, Talstar® and Termidor®. Effect thresholds were determined for survival, growth and swimming performance after short-term (24 h) exposure to insecticides. Results show that both formulation products were more toxic than their pure active ingredients. Detrimental effects on swimming performance were observed at 50% (142 $\mu g.L^{-1}$ fipronil) and 20% (0.14 $\mu g.L^{-1}$ bifenthrin) of the 24-h LC10. Detrimental effects on 7-d growth were observed following 24 h exposure to 53 $\mu g.L^{-1}$ (10% LC10) fipronil.

Observed responses in a suite of biomarker genes demonstrate stress-related cellular effects in exposed fathead minnow larvae at 0.07 (bifenthrin) and 53 (fipronil) µg/L (Beggel et al., Chapter V.7). The initial response at very low concentrations likely indicates disturbance in cell homeostasis leading, with increasing concentration, to adaptive and compensatory responses. Effect concentrations for swimming abnormalities correspond to the onset of the second phase molecular responses, which tend to be dose-dependent. Recovery from insecticide-induced stress was reflected in expression levels of ASPA. Comparisons of biomarker expression data using “heatmaps” were able to distinguish between chemical type and exposure concentration, demonstrating the significant promise these tools hold for toxicity monitoring in field surveys.

II. 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 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 (Sommer 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.

The goal of this study was to assess the role of chemical contaminants in the observed decline of pelagic species in the Delta. We approached this goal from multiple angles, including (i) monitoring of water column toxicity using single-species tests with the amphipod *Hyalella azteca*; (ii) laboratory toxicity tests of ambient samples using larval delta smelt (*Hypomesus transpacificus*); (iii) *in situ* toxicity tests using fathead minnow (*Pimephales promelas*), *H. azteca* and delta smelt; (iv) determination of comparative sensitivity to select contaminants of model species and important resident species; and (v) the development of sensitive biomarker tools for delta smelt and fathead minnow.

Toxicity monitoring with Hyalella azteca: The design of this study built on results of our 2006-2007 Delta-wide monitoring project to investigate toxicity of Delta water samples (Werner et al., 2008, Werner et al., in press). From January 1, 2006 to December 31, 2007, significant amphipod mortality was observed in 5.6 % of ambient samples. Addition of PBO significantly affected survival or growth in 1.1 % and 10.1 % of ambient samples, respectively. Sites in the Lower Sacramento River had the largest number of acutely toxic samples, high occurrence of PBO effects on amphipod growth (along with sites in the South Delta), and the highest total ammonia/um concentrations (0.28 ± 0.15 $mg.L^{-1}$). It was concluded that ammonia/um and/or contaminants occurring in mixture with these, likely contributed to the observed toxicity. Pyrethroid insecticides were detected at potentially toxic concentrations. The 2008-10 study intensified toxicity testing in some ecologically important areas (Cache Slough/lower Sacramento, Suisun Marsh and Bay) of the Delta where acute toxicity was detected in 2007. Water samples were collected bi-weekly from 16 sites located in large delta channels and main-stem rivers, selected based on prevalent distribution patterns of fish species of concern. Ten-day laboratory tests with *H. azteca* survival and relative growth as toxicity endpoints were conducted. The enzyme inhibitor, piperonyl butoxide (PBO, 25 $\mu g.L^{-1}$) was added to synergize or antagonize pyrethroid or organophosphate (OP) insecticide toxicity, respectively. When acute toxicity to the amphipod *H. azteca* ($\geq$50 % mortality within 7 d) or significant PBO effects were detected, toxicity identification evaluations and/or chemical analysis were used in order to identify toxicant(s). If a sample causes $\geq$50 % mortality within 96 h, follow-up samples were collected in an attempt to identify the sources of toxicity. Appropriate sites for follow-up sampling were determined early in 2008 using land use and point source information. In addition, laboratory toxicity tests with larval delta smelt were performed in late April-July on water samples from select locations of special ecological interest such as Cache Slough, lower Sacramento and San Joaquin Rivers, and Suisun Marsh.

Toxicity monitoring with larval delta smelt (H. transpacificus): Toxicity tests using surrogate species are valuable first tier assessments, however, because of their limitations such tests should only be used as a first tier investigation. Sources of uncertainty when extrapolating from surrogate species tests to ecological effects include: variation among species and different life-stages in sensitivity to

chemical stressors, and effects of exposure duration and scenario. This study addressed the above limitations by developing laboratory toxicity tests for one of the resident species of special interest, the delta smelt, and by exposing multiple species in the field, using specially designed *in situ* systems. These *in situ* tests were conducted with fish (delta smelt and fathead minnows) and the invertebrate *H. azteca* at suitable locations (Hood, Rough & Ready Island) in the Delta.

Comparative sensitivity of aquatic species to chemical stressors: The lack of information on the toxic effects of contaminants on resident Delta species, among them delta smelt and important copepod prey species, so far prevents an estimation of the risk of chemical contamination to pelagic organisms of concern. This study addressed an urgent need for information on the comparative sensitivity of these ecologically important species relative to standard test species. Effect concentrations in the form of 96-h LC50, EC50, no observed effect level (NOEC), and lowest observed effect level (LOEC) were generated for several important resident species as well as standard test species. Testing included the copepod, *Eurytemora affinis,* the waterflea*, Ceriodaphnia dubia*, *H. azteca*, delta smelt, and fathead minnow. For standard test species, experiments were performed using laboratory control water as well as Delta water ("hatchery water") to ensure environmental relevance of the test results. Delta smelt was tested in Delta (hatchery) water only. Copepods were tested in laboratory control water. The chemicals were selected based on their known presence in the Delta, recent past or present. They are copper, ammonia, the organophosphate (OP) insecticides chlorpyrifos and diazinon, and the pyrethroid insecticides cyfluthrin, bifenthrin, and permethrin (diazinon was replaced with esfenvalerate for fish testing). Copper is used as a pesticide in various forms, is a common chemical in stormwater runoff, and is ubiquitous in the aquatic environment. Ammonia is released from wastewater treatment plants. Chlorpyrifos is one of the most heavily used agricultural insecticides, and has recently been shown to be present at toxic concentrations in Ulatis Creek (Werner & Kuivila, 2004, unpublished data) and agricultural drains (California Regional Water Quality Control Board Agricultural Waiver Program, 2007). Diazinon, cyfluthrin, bifenthrin and permethrin were detected in 2007 in water column samples from various sites in the Delta (Werner et al., 2008). Bifenthrin has also commonly been detected in sediment samples from the region (K. Larsen, CVRWQCB, personal communication).

Development of biomarker tools for resident fish species of special interest: Sublethal effects of aquatic contaminants are difficult to detect, quantify and interpret in an ecological context. Traditional toxicity tests cannot detect the chronic, sublethal effects of modern contaminants, e.g. endocrine disrupting chemicals, immune suppressants and others, whose effects can have far-reaching deleterious consequences for fish and invertebrate populations in the wild. Approaches involving sensitive and mechanism-based biomarkers of toxic effects therefore need to be developed for future integration in monitoring programs to assess organism health and identify sublethal contaminant effects in fish species of concern. 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 organisms, and can provide information on the causative agents. Molecular biomarkers have been developed for striped bass (Geist et al., 2007, Chapter V), and for delta smelt (Chapter V). A complementary study focused on linking cellular biomarker responses detected in delta smelt and striped bass to ecologically relevant effects such as swimming ability, growth and survival using a model species, the fathead minnow (Chapter V).

III. Toxicity Monitoring in the Sacramento-San Joaquin Delta

III.1. Study Design

III.1.1 Sampling Sites

Water samples for ambient toxicity testing with *H. azteca* were collected bi-weekly from 1 January, 2008 to 31 December, 2009 from 16 sites in the Delta (Tables 1, 2, Figure 1). Water samples for tests with larval delta smelt were collected during spring and early summer from six sites: Suisun, Light 55, Hood, Cache-Lindsey, Rough and Ready and Napa.

Table 1. Site locations and sampling schedule during the 2008-20010 monitoring period.

Station	Location	Latitude	Longitude	Collection Day
340	Napa River, at the seawall	38-05'-51"N	122-15'-43.9"W	Wednesday
405	Carquinez Straight, just west of Benicia army dock	38-02'-22.9"N	122-09'-01.8"W	Wednesday
Suisun [DS]	Suisun Slough at Rush Ranch	38-12'-28.2"N	122-01'56.9"W	Tuesday
508	Suisun Bay, off Chipps Island, opposite Sac. North Ferry Slip	38-02'-43.8"N	121-55'-07.7"W	Wednesday
602	Grizzly Bay, northeast of Suisun Slough at Dolphin	38-06'-50.4"N	122-02'-46.3"W	Wednesday
609	Montezuma Slough at Nurse Slough	38-10'-01.9"N	121.56'-16.8"W	Wednesday
711	Sacramento River at the tip of Grand Island	38-10'-43.7"N	121-39'-55.1"W	Thursday PM
Light 55 [DS]	Sacramento River Deep Water Channel at Light 55	38-16'-26.5"N	121-39'-13.6"W	Thursday AM
Hood [DS]	DWR water quality monitoring station	38-22'-03.6"N	121-31'-13.6"W	Tuesday
Cache-Lin [DS]	Confluence of Lindsey Slough/Cache Slough	38-14'-39.2"N	121-41'-19.5"W	Thursday AM
Cache-Ulatis	Upper Cache Slough, mouth of Ulatis Creek	38-17'-02.7"N	121-43'-04.3"W	Thursday AM
815	San Joaquin, Confluence of Potato Slough	38-05'-06.4"N	121-34'-20.4"W	Thursday PM
902	Old River at mouth of Holland Cut	38-01'-09.1"N	121-34'-55.9"W	Thursday PM
915	Old River, western arm at Railroad Bridge	37-56'-33"N	121-33'-48.6"W	Thursday PM
R&R [DS]	San Joaquin, Rough & Ready Island	37-57'45.4"N	121-21'55.9"W	Tuesday
Napa [DS]	Napa River, Napa City at end of River Park Blvd.	38-16'-39.7"N	122-16'-56.9"W	Tuesday

[DS] Sites sampled for both *H. azteca* and larval delta smelt tests.

Table 2. Follow-up sampling locations during the 2008-2010 monitoring period.

Station	Location	Follow-up Sampling
340	Napa River, Historic 340 at the seawall	Resample of 340
405	Carquinez Straight, just west of Benicia army dock	Resample of 405; Pacheco Creek
Suisun	Suisun Slough, downstream of Boynton Slough	Resample of Suisun; Upstream Boynton Slough, upstream Rush Ranch
508	Suisun Bay, off Chipps Island, opposite Sac. North Ferry Slip	Resample of 508; upstream Sac River, upstream San Joaquin River, 602
602	Grizzly Bay, northeast of Suisun Slough at Dolphin	Resample of 602; Suisun, 609, 508, 405
609	Montezuma Slough at Nurse Slough	Resample of 609; Nurse Slough, Mouth at Van Sickle Island
711	Sacramento River at the tip of Grand Island	Resample of 711; 704, Sac River near Locke, Gate from Moklumne
Light 55	Sacramento River Deep Water Channel at Light 55	Resample of Light 55
Hood	DWR water quality monitoring station	Resample of Hood
Cache-Lin	Confluence of Lindsey Slough/Cache Slough	Resample of Cache-Lin; Lindsey Slough, Cache-Ulatis
Cache-Ulatis	Upper Cache Slough, mouth of Ulatis Creek	Resample of Cache-Ulatis; upstream Ulatis Creek
815	San Joaquin, Confluence of Potato Slough	Resample of 815; Mokelumne Slough, Potato Slough, upstream San Joaquin River, San Joaquin River to Franks Tract Connector, 812
902	Old River at mouth of Holland Cut	Resample of 902; 815, 915, Connection Slough
915	Old River, western arm at Railroad Bridge	Resample of 915; North Woodward Island, 902, Rock Slough
R&R	San Joaquin, Rough & Ready Island	Resample of R&R; Calaveras, Port of Stockton, upstream San Joaquin River, French Camp
Napa	Napa River in Napa City at end of River Park Blvd.	Resample of Napa

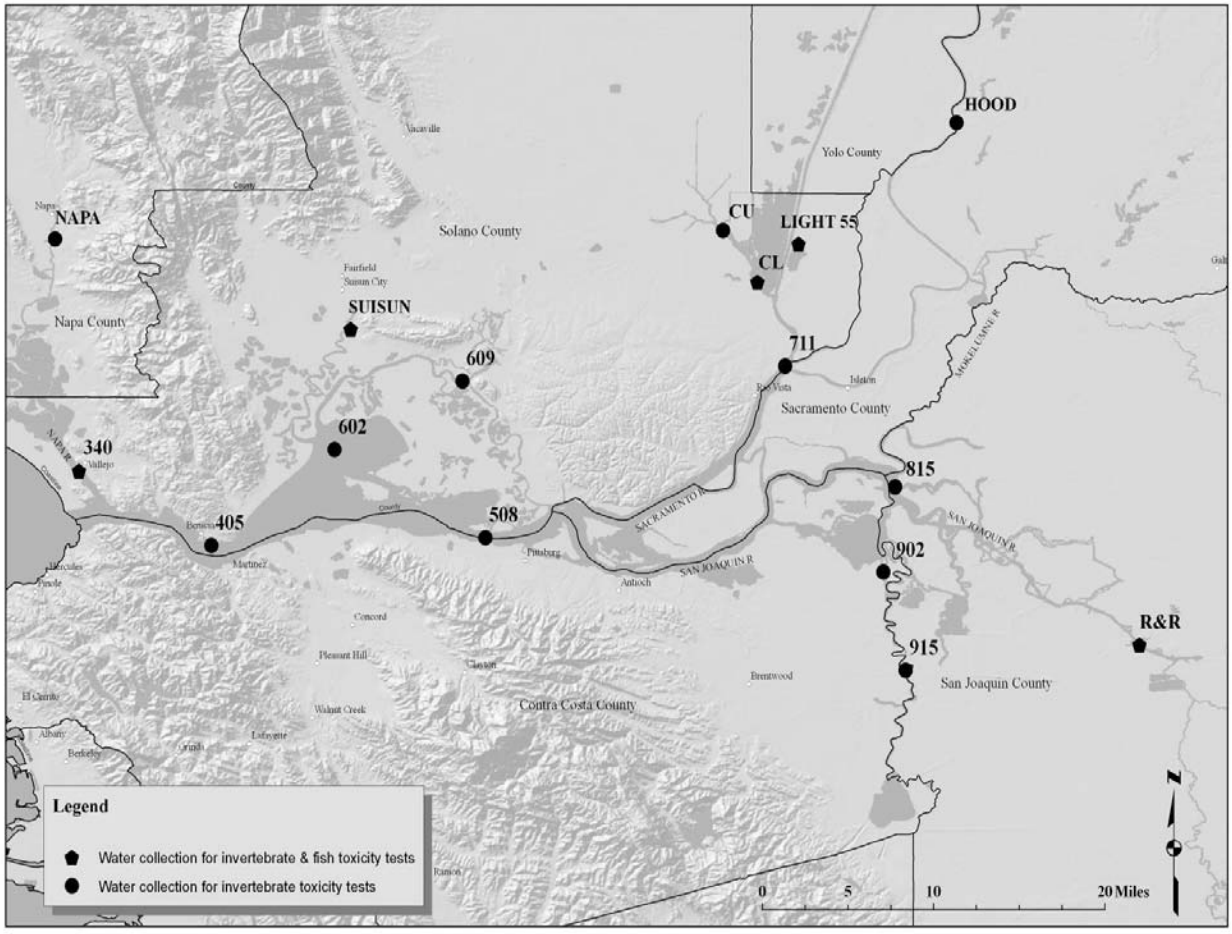

Figure 1. Water sampling locations in 2008-2009, selected based on Interagency Ecological Program summer townet survey stations.

III.1.2 Collection of Water Samples

Staff from the UC Davis Aquatic Toxicology Laboratory (UCD ATL) and the California Department of Fish and Game (CDFG) collected water samples by boat from the following sites: 340, 405, 508, 602, 609, 711, Light 55, Cache-Lindsey, Cache-Ulatis, 815, 902, and 915. Water samples from Hood, Rough & Ready, Suisun, and Napa were collected from shore. If the salinity exceeded the *H. azteca* tolerance limit of 15 ppt, samples were not collected. Subsurface grab samples were pumped from a depth of approximately 0.5 m using a standard water pump into clean, 1-gal amber LDPE cubitainers for invertebrate tests and 5-gal clear LDPE cubitainers for delta smelt tests.

In addition, site water was also collected in 1-gal clear LDPE cubitainers and 1-L amber-glass bottles for analytical chemistry. Water samples were transported, stored and preserved following

protocols outlined in the UCD ATL standard operating procedures (SOP), nos. 5-1 and 5-2 (UCD ATL, 2009). All cubitainers used for water collections were labeled with the site ID, collection date and time, and the initials of the sampler and then rinsed three times with ambient sample water prior to filling. Eight gallons of water were collected from each site for invertebrate testing along with two liters for analytical chemistry. During the spring, an additional 35 gallons were collected from selected sites for delta smelt toxicity testing. All samples were placed into an ice chest on wet ice for transport to the UCD ATL and ice was renewed as needed to keep the sample temperature at 0-6ºC (USEPA, 2002). Upon receipt at UCD ATL, water samples were stored in an environmental chamber at 0-6ºC.

III.1.3 Field Water Quality

Field water quality 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. Meters for DO/SC/EC and pH measurements 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 and a Hach AmVer Ammonia Test'N Tube Reagent Set, respectively. 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, field temperature, field EC and field pH. General weather conditions and GPS coordinates were recorded for each site and sampling event. Tables 3-3 and 3-4 summarize minimum and maximum water quality data by site.

III.2. Monitoring with *Hyalella azteca*

III.2.1. Methods

III.2.1.1 10-Day Acute and Chronic Water Column Toxicity Tests

H. azteca were purchased from Aquatic Research Organisms (Hampton, NH, USA). Upon receipt, amphipods were moved to 10-L aquaria, fed, and acclimated to laboratory test conditions for 48 h. The 10-d testing procedure used in this study was based on protocols described in the Quality Assurance Management Plan for the State of California's Surface Water Ambient Monitoring Program (California State Water Resources Control Board, http://www.waterboards.ca.gov/water_issues/programs/swamp/qamp.shtml#appendixf). At test initiation, water samples were shaken vigorously in original sampling containers, and sub-samples were filtered through a 53-µm screen to remove debris and larger organisms. Water was then warmed to test temperature (23 ± 1°C) in 600 ml beakers using a water bath maintained at 25±2°C, and aerated at a rate of 100 bubbles per minute until DO concentration was 4.9-8.9 $mg.L^{-1}$. De-ionized water amended to US EPA moderately hard specifications (hardness: 90-100 $mg.L^{-1}$ $CaCO_3$, alkalinity: 50-70 $mg.L^{-1}$ $CaCO_3$, electrical conductivity (EC): 330-360 $\mu S.cm^{-1}$, pH, 7.8-8.2; US EPA, 2002) was used for controls. One or multiple high-conductivity controls were added when the SC of an ambient sample was >10,000 $\mu S.cm^{-1}$. A low conductivity control was added when the SC of an ambient sample was <100 $\mu S.cm^{-1}$. Filtered (1 µm A/E glass fiber filter) Pacific Ocean seawater from Bodega Bay Marine Laboratory, Bodega Bay, CA, or de-ionized water was used to increase or decrease the conductivity of control water.

Tests were initiated with 9-14 d old *H. azteca*. Each of four replicate 250-ml glass beakers contained 100 ml of water, a small piece of nitex screen (approx. 6 cm^2) for use as substrate for *H.*

azteca, and 10 organisms. Animals were fed a mixture of yeast, organic alfalfa and trout chow (1 ml per replicate) at test initiation and on days 2, 4, 5, 6, and 8 after water renewal. Tests were conducted at 23 ± 2°C with a 16h:8h L:D photoperiod. Mortality was recorded daily, and 80% of water was renewed every second day. On day 10, the surviving organisms were dried to constant weight at 103-105 °C, and weighed using a Mettler AE 163 balance.

Because the test protocol was changed from one water renewal on day 5 in our 2006-2008 monitoring study, to four water renewals on days 2, 4, 6 and 8 during the 2008-2010 study presented here, a methods comparison test was conducted at the beginning of the project to determine which method provided the greatest sensitivity to contaminants. Three different methods were tested concurrently using permethrin concentrations ranging from 30 to120 ng/L. Methods tested included the following: 1) day 5 renewal with 250 ml beakers; 2) every other day renewal with 250 ml beakers; and 3) every other day renewals with 20 ml scintillation vials. The 250 ml beakers contained 100 ml of sample and 10 organisms per replicate, whereas the scintillation vials contained 15 ml of sample and five animals per replicate. One-inch square piece of nitex screen were added to each test container as artificial substrate. Organisms were fed YCT at 1000 µl per 250 ml beaker or 150 µl per scintillation vial, on days 0, 2, 4, 6, and 8, regardless of water renewal frequency. Results showed that the 10-day LC_{50} was lowest for the "every other day renewal method" compared to the other two methods (Table 3). This most sensitive method was used as of January 1, 2008, for the entire duration of the 2008-2010 monitoring project.

Table 3. Effect concentrations derived from three *H. azteca* test methods using permethrin as toxicant.

Method	Endpoint	NOEC ng/L	LOEC ng/L	LC50 ng/L
Day 5 Renewal – 250 ml beakers	96 h	30	60	57.9
Day 5 Renewal – 250 ml beakers	10 d	30	60	53.8
Every other day Renewal – 250 ml beakers	96 h	30	60	53.7
Every other day Renewal – 250 ml beakers	10 d	30	60	36.2
Every other day Renewal –20 ml scintillation vials	96 h	30	60	50.8
Every other day Renewal – 20 ml scintillation vials	10 d	30	60	46.8

Addition of PBO: All tests were conducted with and without PBO to synergize (Amweg and Weston, 2007) or antagonize (Bailey et al., 1996) toxicity of pyrethroid or OP insecticides, respectively, due to concerns that toxicity may be lost during sampling, transport and storage, and to guide subsequent chemical analyses. Tests were conducted with 25 $\mu g.L^{-1}$ PBO. This concentration does not affect amphipod survival or growth (Werner et al., in press).

III.2.1.2 Toxicity Identification Evaluation

Phase I toxicity identification evaluations (TIEs) involve procedures to either remove or inactivate specific classes of chemicals (US EPA 1991). In this study, phase I TIEs were conducted on samples that caused at least 50% mortality within 7 d. Samples that met these criteria were collected at site "711" (Sacramento River at Rio Vista) on June 25, 2009, and at site "Suisun" (Suisun Slough) on August 12, 2008. Additional, pyrethroid-focused TIEs were conducted on two samples that showed significantly reduced survival after PBO addition compared to the ambient sample. These were two samples collected from site "Cache-Ulatis" on January 31, 2008 and October 15, 2009.

Toxicity identification evaluation treatments included: air stripping (aeration at 150 bubbles per

min for 2 h) to reduce or remove toxicity due to volatile chemicals such as surfactants, chlorine and ammonia; low test temperature (15^{o}C) to increase toxicity due to pyrethroid insecticides; addition of EDTA (3 concentrations) to chelate metals, making them unavailable to biota; addition of PBO (25 µg.L^{-1}, see above); (v) removal of non-polar organic chemicals by solid phase extraction columns (Varian Bond Elut C8, Varian Inc., Harbor City, CA, USA). Appropriate control and method blank treatments were included for all TIE manipulations. Improved organism performance after TIE manipulation is defined as the absence or a delay of mortality by greater than or equal to 24 h.

III.2.1.3 Statistical Analysis

All statistical analyses were performed using JMP 5.0.1 (SAS Institute Inc., Cary, NC, USA; 1989-2003). Survival and final weight data obtained for ambient samples were compared to their PBO-containing counterpart, and to controls using the U.S. Environmental Protection Agency standard statistical procedures for single concentration static renewal toxicity tests (US EPA, 2002). Shapiro-Wilk's test and Bartlett's test were used to examine normality of distributions and homogeneity of variances (alpha = 0.01). Each sample was compared individually to the appropriate electrical conductivity (EC) control. A one-tailed Wilcoxon (Mann-Whitney) test was used (alpha = 0.05) when data distribution was non-normal in either treatment. When distributions were normal, a homoschedastic or heteroschedastic one-tailed t-test was performed (alpha= 0.05), depending on the presence or absence of homogeneity of variance. Comparisons between sample and sample with PBO used the same approach with two-tailed tests (alpha= 0.05).

Field ammonia/um site-by-site analysis and effects on *H. azteca*: Data of measured total ammonia/um and un-ionized ammonia concentrations were analyzed in more detail. Total ammonia/um as well as unionized ammonia "field" concentrations (calculated using pH, SC and temperature measured in the field) were compared between sampling sites using ANOVA with Tukey's multiple comparison procedure ($p \leq 0.05$). The relationships between ammonia/um in ambient water samples (calculated using pH, EC and temperature measured at test initiation) and *H. azteca* survival and final weight were examined using multivariate regression models with linear and quadratic terms for EC effects, and ammonia/um concentration as a linear effect. Models were examined using data from each site individually as well as from all sites combined.

III.2.1.4 Analytical Chemistry

Water samples for analytical chemistry were collected at each sampling site during each sampling event using two acid-cleaned, 1-L amber-glass bottles. These samples were transported on ice and stored in an environmental chamber maintained at 4^{o}C upon receipt at the UCD ATL. Samples for organic chemical analysis were preserved by addition of 10 ml/l dichloromethylene immediately upon receipt at the laboratory due to concerns that labile organic chemicals could break down during storage. Samples were submitted to the California Department of Fish and Game Water Pollution Laboratory, Rancho Cordova, CA for organic chemical analysis. Samples submitted for total and/or dissolved metals analysis were analyzed at the California Department of Fish and Game – Moss Landing Marine Laboratory, Moss Landing, CA.

Samples that showed significant differences in survival or growth between PBO-treated and ambient samples based on US EPA standard statistics were analyzed for either pyrethroids (bifenthrin, cyfluthrin, cypermethrin, deltamethrin, esfenvalerate/fenvalerate, fenpropathrin, lambda-cyhalothrin, cis/trans permethrin) and/or 41 OP insecticides (Appendix A, Table A1) by gas chromatography (Agilent 6890 plus, Agilent Technologies Inc., Santa Clara, CA, USA) with dual columns (DB5 and

DB17) and dual flame photometric detectors in phosphorous mode (OP insecticides), or dual micro-electron capture detectors (pyrethroid insecticides). Pyrethroids were confirmed using GC-MS or GC-MSMS. When the possible cause of toxicity was less apparent, water samples were analyzed for a "comprehensive" suite of chemicals including metals (dissolved and total), PAHs, pyrethroids, organophosphates, carbamates, and fipronil and degradates following standard methods. Analytes and respective detection/reporting limits are provided in Appendix A, Table A1.

III.2.1.5 Quality Assurance/Quality Control

Test acceptability criteria for *H. azteca* toxicity tests required 90% control survival California State Water Resources Control Board, (http://www.waterboards.ca.gov/water_issues/programs/swamp/qamp.shtml#appendixf). To evaluate whether organism sensitivity was consistent throughout the project period, positive control reference toxicant tests were performed once a month using NaCl as the toxicant. If an effect concentration, LC_{50} or EC_{25}, was outside the 95% confidence interval, test organism sensitivity was considered atypical and results of tests conducted during that month were considered suspect. To assess laboratory testing precision, 48 duplicate ambient water samples were collected and tested. In addition, 30 bottle blanks and 24 trip blanks were tested to ascertain the cleanliness of the sampling container, and detect potential contamination of water samples during collection and transport. Any deviations from test protocols were recorded.

III.2.2 Results

Water quality parameters measured at field sites are shown as mean (+/- SD), and maximum-minimum ranges in Table 4 a. Sites were ranked with respect to their ammonia/um concentrations for the study periods 2008-2010 and 2006-2010 (Tables 4 b, c). Sites 711, Cache-Lindsey and Hood had the highest concentrations of ammonia/um.

Table 4 a. Water quality at sites sampled during January 2008 – December 2009.

Year	Site	*N*	SC (uS/cm)		EC (uS/cm)		Temp (°C)	
			Mean ± SD	Range	Mean ± SD	Range	Mean ± SD	Range
2008	340	8	17579 ± 5151	10250 - 23250	13321 ± 4325	7470 - 18510	12.0 ± 2.3	8.7 - 15.1
	405	15	16688 ± 4315	8980 - 23070	13728 ± 4481	6750 - 20840	15.0 ± 4.0	8.5 - 20.2
	508	26	6244 ± 3875	386.2 - 13000	5221 ± 3115	272.8 - 10550	15.8 ± 4.6	7.8 - 22.2
	602	26	13447 ± 6799	364.4 - 22140	11347 ± 5862	262.3 - 18580	16.0 ± 4.3	8.3 - 21.8
	609	26	9204 ± 3996	3497 - 15420	7805 ± 3661	2559 - 13900	16.3 ± 4.8	8.4 - 23.2
	711	25	218 ± 66	146 - 385.7	181 ± 48	101.1 - 311.1	16.6 ± 5.7	7.5 - 24.5
	815	25	328 ± 101	217.7 - 722	273 ± 70	177.9 - 488	16.8 ± 5.6	7.9 - 24.9
	902	25	579 ± 292	293.4 - 1444	481 ± 226	218.9 - 1030	16.8 ± 6.0	6.9 - 24.9
	910	1	610	-	406	-	7.4	-
	915	25	519 ± 159	315.2 - 888	430 ± 123	238 - 656	17.2 ± 6.1	6.9 - 25.3
	CL	26	262 ± 75	167.2 - 459.4	214 ± 53	159.1 - 371.9	16.3 ± 5.3	7.6 - 25.0
	CU	26	389 ± 160	197.1 - 826	313 ± 116	182.5 - 668	16.0 ± 5.1	6.8 - 24.7
	Hood	26	214 ± 58	151.8 - 392.8	182 ± 48	124.1 - 291.1	17.6 ± 5.6	7.9 - 27.7
	Light 55	25	304 ± 56	212.2 - 436.2	253 ± 50	172.2 - 392.2	16.2 ± 5.3	7.9 - 23.6
	Napa	16	9827 ± 9291	260 - 25790	8717 ± 8604	199.7 - 24965	16.3 ± 4.5	9.2 - 23.4
	R&R	25	710 ± 161	398.6 - 1045	626 ± 147	282.2 - 883	19.1 ± 5.8	9.0 - 29.8
	Suisun @ RR	24	9993 ± 4765	3089 - 16070	8411 ± 4360	2391 - 15320	16.2 ± 4.3	7.1 - 24.5
	Suisun Pub. Dock	2	4424 ± 2173	2887 - 5960	3006 ± 1322	2071 - 3941	8.9 ± 2.3	7.3 - 10.5
2009	340	9	18661 ± 5367	9460 - 24360	15117 ± 4660	7100 - 20060	14.8 ± 2.9	10.0 - 18.6
	405	17	17559 ± 5934	6510 - 24290	14932 ± 5356	4880 - 22110	16.8 ± 3.2	10.2 - 20.6
	508	26	6609 ± 3772	358.3 - 12810	5299 ± 2720	279 - 9260	15.6 ± 4.5	8.3 - 21.7
	602	26	13934 ± 6629	424.9 - 23550	11399 ± 5209	333 - 17440	15.9 ± 4.5	8.6 - 22.1
	609	25	8221 ± 4381	2229 - 15450	6887 ± 3701	1711 - 12960	16.6 ± 4.8	9.2 - 22.8
	711	26	191 ± 69	110.7 - 416.5	156 ± 48	100.6 - 302.6	16.4 ± 5.3	7.9 - 23.2
	815	26	300 ± 122	159.8 - 572	247 ± 89	145.5 - 433	17.0 ± 5.4	7.8 - 23.3
	902	26	507 ± 236	204 - 854	422 ± 192	194.3 - 835	17.3 ± 5.7	7.6 - 24.5
	915	26	487 ± 193	216.8 - 846	410 ± 162	209 - 834	17.6 ± 6.0	7.6 - 25.1
	CL	26	241 ± 65	154.6 - 417.3	197 ± 54	142 - 388	15.9 ± 4.8	7.9 - 22.1
	CU	26	363 ± 157	135.6 - 674	288 ± 109	122.6 - 545	15.6 ± 4.7	7.3 - 21.2
	Hood	25	184 ± 41	116.3 - 303.3	151 ± 28	100.7 - 220	16.5 ± 5.2	8.2 - 23.9
	Light 55	26	292 ± 53	214.5 - 408.8	241 ± 46	172.5 - 342	16.2 ± 5.1	7.9 - 22.0
	Napa	20	12696 ± 8610	237.2 - 24240	10212 ± 6772	178.4 - 19780	15.7 ± 4.7	9.6 - 23.9
	R&R	25	694 ± 218	395.3 - 1107	580 ± 127	322.5 - 811	18.2 ± 6.4	8.2 - 26.9
	Suisun @ RR	25	8952 ± 3705	2673 - 14660	7049 ± 2847	2038 - 12100	15.2 ± 4.3	7.8 - 22.1

Table 4 a, continued. Water quality at sites sampled during January 2008 – December 2009.

Year	Site	*N*	pH		DO (mg/L)		Turbidity (NTU)	
			Mean ± SD	Range	Mean ± SD	Range	Mean ± SD	Range
2008	340	8	7.75 ± 0.32	7.09 - 8.03	10.5 ± 0.4	10.0- 11.2	41 ± 23	13 - 74
	405	15	7.66 ± 0.41	6.43 - 8.05	10.1 ± 1.0	8.1 - 11.7	132 ± 109	12 - 378
	508	26	7.67 ± 0.33	6.80 - 8.24	10 ± 1.1	8.2 - 12.1	28 ± 26	7 - 120
	602	26	7.77 ± 0.27	6.95 - 8.28	9.9 ± 1.1	8.0 - 12.3	61 ± 74	9 - 306
	609	26	7.49 ± 0.33	6.51 - 8.04	9.3 ± 1.1	7.4 - 11.6	35 ± 24	8 - 103
	711	25	7.48 ± 0.38	6.82 - 8.16	9.9 ± 1.8	7.2 - 14.5	27 ± 52	4 - 243
	815	25	7.58 ± 0.42	6.75 - 8.31	9.9 ± 1.6	7.6 - 13.8	13 ± 17	3 - 80
	902	25	7.58 ± 0.44	6.59 - 8.30	10 ± 1.8	7.8 - 14.9	20 ± 35	3 - 184
	910	1	7.55	-	12.5	-	6	-
	915	25	7.60 ± 0.39	6.78 - 8.20	9.8 ± 1.9	7.5 - 14.8	11 ± 8	4 - 44
	CL	26	7.61 ± 0.4	6.80 - 8.29	9.9 ± 1.7	7.5 - 14.4	44 ± 41	7 - 217
	CU	26	7.80 ± 0.44	6.76 - 8.43	10.1 ± 1.7	8.3 - 15.1	54 ± 30	8 - 119
	Hood	26	7.42 ± 0.32	6.80 - 8.05	9 ± 1.5	7.1 - 11.7	20 ± 26	5 - 107
	Light 55	25	7.64 ± 0.37	7.08 - 8.36	9.8 ± 1.3	8.1 - 12.5	51 ± 45	8 - 240
	Napa	16	7.66 ± 0.48	6.96 - 8.70	9.1 ± 2.4	5.4 - 12.4	45 ± 46	7 - 195
	R&R	25	7.52 ± 0.31	6.92 - 8.10	8.4 ± 1.3	5.8 - 11.1	14 ± 9	5 - 45
	Suisun @ RR	24	7.35 ± 0.28	6.74 - 7.97	7.5 ± 1.6	5.1 - 11.0	62 ± 45	15 - 177
	Suisun Pub. Dock	2	7.48 ± 0.25	7.30 - 7.65	9.3 ± 1.3	8.4 - 10.2	161 ± 149	56 - 266
2009	340	9	7.23 ± 0.45	6.58 - 7.88	10 ± 1.3	8.7 - 13.0	52 ± 21	21 - 78
	405	17	7.49 ± 0.24	6.99 - 7.89	9.6 ± 1.2	8.1 - 13.0	112 ± 115	11 - 424
	508	26	7.46 ± 0.32	6.54 - 7.87	9.9 ± 1.0	8.5 - 11.8	22 ± 13	8 - 49
	602	26	7.58 ± 0.30	6.75 - 8.00	10 ± 1.2	8.6 - 13.4	74 ± 90	8 - 379
	609	25	7.33 ± 0.28	6.66 - 7.70	9.1 ± 1.4	7.4 - 11.9	45 ± 34	12 - 138
	711	26	7.43 ± 0.36	6.61 - 8.60	9.6 ± 1.3	8.1 - 12.1	17 ± 31	3 - 146
	815	26	7.43 ± 0.36	6.58 - 8.55	9.6 ± 1.3	8.2 - 12.0	5 ± 3	2 - 17
	902	26	7.59 ± 0.40	6.58 - 8.58	9.7 ± 1.3	8.0 - 12.1	6 ± 4	2 - 19
	915	26	7.51 ± 0.39	6.43 - 8.61	9.5 ± 1.5	7.7 - 12.6	5 ± 2	2 - 11
	CL	26	7.53 ± 0.33	6.80 - 8.59	9.9 ± 1.2	8.4 - 12.1	30 ± 25	6 - 133
	CU	26	7.70 ± 0.40	6.88 - 8.61	9.8 ± 1.7	6.4 - 13.8	56 ± 43	9 - 164
	Hood	25	7.29 ± 0.28	6.55 - 7.67	9.4 ± 1.3	7.5 - 12.0	13 ± 8	5 - 44
	Light 55	26	7.67 ± 0.36	7.02 - 8.38	9.7 ± 1.2	8.3 - 12.5	27 ± 18	8 - 97
	Napa	20	7.43 ± 0.34	6.51 - 7.98	9.5 ± 1.7	6.1 - 11.4	64 ± 151	4 - 713
	R&R	25	7.51 ± 0.24	7.08 - 7.94	8.5 ± 1.9	5.6 - 11.3	7 ± 3	2 - 13
	Suisun @ RR	25	7.23 ± 0.22	6.51 - 7.53	8.3 ± 1.7	5.5 - 11.9	63 ± 93	9 - 395

Table 4 a, continued. Water quality at sites sampled during January 2008 – December 2009.

Year	Site	*N*	Hardness (mg/L as CaCO3)		Alkalinity (mg/L as CaCO3)	
			Mean ± SD	Range	Mean ± SD	Range
2008	340	8	1976 ± 593	1000 - 2640	100 ± 12	82 - 116
	405	15	1798 ± 658	224 - 2720	99 ± 6	88 - 112
	508	26	702 ± 465	60 - 1560	87 ± 8	70 - 101
	602	26	1478 ± 802	96 - 2560	94 ± 7	76 - 106
	609	26	1132 ± 530	408 - 2320	99 ± 11	80 - 114
	711	25	76 ± 28	52 - 200	74 ± 12	53 - 98
	815	25	83 ± 14	60 - 116	82 ± 28	64 - 212
	902	25	109 ± 22	76 - 156	77 ± 8	60 - 88
	910	1	120	-	88	-
	915	25	104 ± 18	52 - 132	81 ± 19	66 - 166
	CL	26	84 ± 19	56 - 128	87 ± 17	62 - 126
	CU	26	122 ± 53	44 - 248	116 ± 38	70 - 222
	Hood	26	67 ± 15	28 - 92	73 ± 12	48 - 98
	Light 55	25	98 ± 16	76 - 132	93 ± 15	66 - 124
	Napa	16	1227 ± 1151	76 - 3160	115 ± 31	62 - 148
	R&R	25	154 ± 35	88 - 244	97 ± 17	64 - 126
	Suisun @ RR	24	1282 ± 642	400 - 2440	167 ± 15	136 - 192
	Suisun Pub. Dock	2	578 ± 269	388 - 768	187 ± 30	166 - 208
2009	340	9	2177 ± 675	1040 - 2880	98 ± 6	88 - 108
	405	17	1952 ± 657	800 - 2840	89 ± 6	80 - 104
	508	26	788 ± 514	92 - 2120	78 ± 13	56 - 106
	602	26	1643 ± 786	152 - 3040	86 ± 11	68 - 106
	609	25	964 ± 518	292 - 1760	93 ± 8	78 - 116
	711	26	67 ± 16	44 - 100	66 ± 15	46 - 96
	815	26	80 ± 20	48 - 124	67 ± 13	50 - 94
	902	26	98 ± 27	56 - 156	70 ± 12	50 - 92
	915	26	100 ± 23	64 - 152	72 ± 12	54 - 98
	CL	26	80 ± 17	54 - 116	80 ± 18	54 - 118
	CU	26	120 ± 49	60 - 226	110 ± 39	62 - 204
	Hood	25	65 ± 15	44 - 100	68 ± 14	50 - 92
	Light 55	26	94 ± 15	64 - 124	87 ± 16	66 - 124
	Napa	20	1585 ± 1032	70 - 2800	124 ± 44	48 - 284
	R&R	25	150 ± 40	92 - 212	97 ± 33	68 - 240
	Suisun @ RR	25	1028 ± 469	160 - 1880	169 ± 34	130 - 274

Table 4 a, continued. Water quality at sites sampled during January 2008 – December 2009.

Year	Site	*N*	Total Ammonia Nitrogen (mg/L)		Unionized Ammonia (mg/L)	
			Mean ± SD	Range	Mean ± SD	Range
2008	340	8	0.15 ± 0.09	0 - 0.27	0.001 ± 0.001	0 - 0.002
	405	15	0.19 ± 0.07	0.07 - 0.31	0.002 ± 0.001	0 - 0.004
	508	26	0.11 ± 0.08	0.01 - 0.29	0.001 ± 0.001	0 - 0.005
	602	26	0.12 ± 0.07	0 - 0.29	0.001 ± 0.001	0 - 0.004
	609	26	0.12 ± 0.10	0 - 0.27	0.001 ± 0.001	0 - 0.003
	711	25	0.37 ± 0.13	0.03 - 0.58	0.005 ± 0.006	0 - 0.021
	815	25	0.12 ± 0.07	0.01 - 0.24	0.002 ± 0.001	0 - 0.006
	902	25	0.08 ± 0.11	0 - 0.51	0.001 ± 0.001	0 - 0.003
	910	1	0.13	-	0.001	-
	915	25	0.07 ± 0.09	0 - 0.44	0.001 ± 0.001	0 - 0.003
	CL	26	0.22 ± 0.10	0.07 - 0.44	0.003 ± 0.003	0 - 0.009
	CU	26	0.10 ± 0.06	0 - 0.20	0.002 ± 0.001	0 - 0.006
	Hood	26	0.38 ± 0.13	0.08 - 0.59	0.004 ± 0.004	0 - 0.019
	Light 55	25	0.16 ± 0.11	0 - 0.44	0.002 ± 0.002	0 - 0.008
	Napa	16	0.11 ± 0.12	0 - 0.37	0.001 ± 0.001	0 - 0.002
	R&R	25	0.12 ± 0.08	0.03 - 0.40	0.001 ± 0.001	0 - 0.006
	Suisun	24	0.13 ± 0.06	0.04 - 0.23	0.001 ± 0.001	0 - 0.003
	Suisun Pub. Dock	2	0.31 ± 0.01	0.30 - 0.32	0.001 ± 0.001	0.001 - 0.002
2009	340	9	0.19 ± 0.16	0.09 - 0.59	0.001 ± 0.001	0 - 0.002
	405	17	0.19 ± 0.14	0 - 0.62	0.001 ± 0.001	0 - 0.003
	508	26	0.14 ± 0.08	0 - 0.31	0.001 ± 0.001	0 - 0.003
	602	26	0.16 ± 0.08	0.03 - 0.33	0.001 ± 0.001	0 - 0.003
	609	25	0.18 ± 0.11	0 - 0.36	0.001 ± 0.001	0 - 0.002
	711	26	0.28 ± 0.15	0.04 - 0.56	0.003 ± 0.004	0 - 0.021
	815	26	0.11 ± 0.08	0 - 0.25	0.001 ± 0.002	0 - 0.011
	902	26	0.05 ± 0.04	0 - 0.12	0.001 ± 0.001	0 - 0.003
	915	26	0.03 ± 0.03	0 - 0.12	0.000 ± 0.000	0 - 0.002
	CL	26	0.19 ± 0.10	0.05 - 0.39	0.002 ± 0.005	0 - 0.025
	CU	26	0.09 ± 0.07	0 - 0.25	0.002 ± 0.002	0 - 0.010
	Hood	25	0.32 ± 0.16	0.02 - 0.55	0.002 ± 0.001	0 - 0.004
	Light 55	26	0.14 ± 0.09	0.03 - 0.33	0.002 ± 0.003	0 - 0.014
	Napa	20	0.14 ± 0.11	0 - 0.44	0.001 ± 0.001	0 - 0.004
	R&R	25	0.10 ± 0.08	0 - 0.43	0.001 ± 0.001	0 - 0.006
	Suisun	25	0.16 ± 0.10	0 - 0.46	0.001 ± 0.001	0 - 0.004

Table 4 b. Ranking of sites with respect to total ammonia/um and un-ionized ammonia concentrations at sampling sites, 2008-2009. Un-ionized ammonia concentrations were calculated using water temperature, EC and pH measured at the time of sampling. Sites are ranked with different letters to indicate statistical differences ($p \leq 0.05$), where A represents sites with highest and E or F represents sites with lowest concentrations.

	Total Ammonia Nitrogen (mg/L)				Unionized Ammonia (mg/L)			
Site	*N*	Mean ± SD	Range	Rank	*N*	Mean ± SD	Range	Rank
340	17	0.17 ± 0.13	0 - 0.59	B C D	17	0.001 ± 0.001	0 - 0.002	D E F
405	32	0.19 ± 0.11	0 - 0.62	B C	32	0.002 ± 0.001	0 - 0.004	B C D E
508	52	0.13 ± 0.08	0 - 0.31	C D E	52	0.001 ± 0.001	0 - 0.005	D E F
602	52	0.14 ± 0.08	0 - 0.33	C D	52	0.001 ± 0.001	0 - 0.004	C D E F
609	51	0.15 ± 0.11	0 - 0.36	B C D	51	0.001 ± 0.001	0 - 0.003	E F
711	**52**	**0.33 ± 0.14**	**0.03 - 0.58**	**A**	**51**	**0.004 ± 0.005**	**0 - 0.021**	**A**
815	51	0.12 ± 0.08	0 - 0.25	D E	51	0.001 ± 0.002	0 - 0.011	D E F
902	51	0.06 ± 0.08	0 - 0.51	E	51	0.001 ± 0.001	0 - 0.003	F
910	1	0.13	-	-	1	0.001	-	-
915	51	0.05 ± 0.07	0 - 0.44	E	51	0 ± 0.001	0 - 0.003	F
CL	**52**	**0.20 ± 0.10**	**0.05 - 0.44**	**B**	**52**	**0.003 ± 0.004**	**0 - 0.025**	**A B C**
CU	52	0.09 ± 0.07	0 - 0.25	D E	52	0.002 ± 0.002	0 - 0.010	C D E F
Hood	**52**	**0.35 ± 0.15**	**0.02 - 0.59**	**A**	**51**	**0.003 ± 0.003**	**0 - 0.019**	**A B**
Light 55	52	0.15 ± 0.10	0 - 0.44	B C D	51	0.002 ± 0.003	0 - 0.014	A B C D
Napa	37	0.13 ± 0.11	0 - 0.44	C D E	36	0.001 ± 0.001	0 - 0.004	E F
R&R	51	0.11 ± 0.08	0 - 0.43	D E	50	0.001 ± 0.001	0 - 0.006	D E F
Suisun	50	0.15 ± 0.08	0 - 0.46	B C D	49	0.001 ± 0.001	0 - 0.004	F
Suisun Pub. Dock	2	0.31 ± 0.01	0.30 - 0.32	-	2	0.001 ± 0.001	0.001 - 0.002	-

Table 4 c. Ranking of sites with respect to total ammonia/um and un-ionized ammonia concentrations at sampling sites, 2006-2009. Un-ionized ammonia concentrations were calculated using water temperature, EC and pH measured at the time of sampling. Sites are ranked with different letters to indicate statistical differences ($p \leq 0.05$), where A represents sites with highest and E sites with lowest concentrations.

	Total Ammonia Nitrogen (mg/L)				Unionized Ammonia (mg/L)			
Site	*N*	Mean ± SD	Range	Rank	*N*	Mean ± SD	Range	Rank
323	14	0.11 ± 0.04	0.06 - 0.20	C D E	14	0.001 ± 0.001	0 - 0.003	A B C D E
340	55	0.11 ± 0.10	0 - 0.59	D E	54	0.001 ± 0.001	0 - 0.002	E
405	79	0.15 ± 0.10	0 - 0.62	C D	79	0.002 ± 0.001	0 - 0.006	B C D E
504	50	0.10 ± 0.06	0 - 0.26	E	50	0.001 ± 0.001	0 - 0.005	E
508	102	0.11 ± 0.07	0 - 0.31	D E	102	0.001 ± 0.001	0 - 0.006	E
602	101	0.12 ± 0.07	0 - 0.33	D E	101	0.001 ± 0.001	0 - 0.005	C D E
609	101	0.14 ± 0.09	0 - 0.36	D E	100	0.001 ± 0.001	0 - 0.003	E
704	50	0.11 ± 0.07	0 - 0.30	D E	50	0.001 ± 0.001	0 - 0.005	C D E
711	**102**	**0.27 ± 0.14**	**0.03 - 0.58**	**B**	**100**	**0.004 ± 0.004**	**0 - 0.021**	**A**
804	50	0.09 ± 0.06	0 - 0.29	E	50	0.001 ± 0.002	0 - 0.008	C D E
812	48	0.09 ± 0.06	0 - 0.29	E	48	0.001 ± 0.001	0 - 0.005	E
815	51	0.12 ± 0.08	0 - 0.25	D E	51	0.001 ± 0.002	0 - 0.011	E
902	101	0.06 ± 0.07	0 - 0.51	E	100	0.001 ± 0.002	0 - 0.010	E
910	51	0.15 ± 0.10	0 - 0.44	C D E	50	0.002 ± 0.002	0 - 0.007	B C D E
915	101	0.06 ± 0.07	0 - 0.44	E	100	0.001 ± 0.001	0 - 0.006	E
CL	**52**	**0.20 ± 0.10**	**0.05 - 0.44**	**C**	**52**	**0.003 ± 0.004**	**0 - 0.025**	**A B C**
CU	53	0.09 ± 0.07	0 - 0.25	E	53	0.002 ± 0.002	0 - 0.010	C D E
Hood	**61**	**0.34 ± 0.14**	**0.02 - 0.59**	**A**	**59**	**0.003 ± 0.003**	**0 - 0.019**	**A B**
Light 55	100	0.13 ± 0.09	0 - 0.44	D E	98	0.002 ± 0.003	0 - 0.014	A B C D
Napa	37	0.13 ± 0.11	0 - 0.44	D E	36	0.001 ± 0.001	0 - 0.004	E
R&R	51	0.11 ± 0.08	0 - 0.43	D E	50	0.001 ± 0.001	0 - 0.006	D E
Stockton WWTF	1	0.21	-	-	1	0.003	-	-
Suisun	50	0.15 ± 0.08	0 - 0.46	C D E	49	0.001 ± 0.001	0 - 0.004	E
Suisun Pub. Dock	2	0.31 ± 0.01	0.30 - 0.32	-	2	0.002 ± 0.001	0.001 - 0.002	-

III.2.2.1 Acute Toxicity to *H. azteca* - Effects on 10-d Survival

During the project period, a total of 752 water samples were collected and tested for toxicity with *H. azteca*. Samples that caused significant mortality, with or without PBO, are listed below in Table 5, along with TIE results and analytical chemistry information. Detailed data tables of all *H. azteca* test results are provided in Appendix B. Four water samples (0.5 % of total) collected from sites 711, Light 55, 405 and Suisun were acutely toxic, causing a significant reduction in amphipod survival within the 10 d test period. Two of these (sites 711, Suisun) had less than 50 % survival, the other two had more than 80 % survival.

PBO Effect on 10-d Survival: The addition of PBO increased acute toxicity in seven ambient samples (0.9 % of total, Table 4-1) suggesting the presence of pyrethroid insecticides. Three of these contained detectable concentrations of pyrethroids: cypermethrin (site 902, Aug. 28, 2008); bifenthrin and lambda cyhalothrin (Cache-Ulatis, Feb. 28, 2008) and permethrin (Hood, Jun. 23, 2009). Survival in six PBO-containing samples (0.8 % of total) was significantly lower than in the respective PBO-controls.

III.2.2.2 Toxicity Identification Evaluation

Samples that were subject to TIEs were collected from site Cache-Ulatis on Jan. 31, 2008 and Oct. 15, 2009, and from Suisun Slough on August 12, 2008 (Table 5). All TIEs indicated that pyrethroid insecticides were the dominant toxicants. However, chemical analyses only detected OP insecticides at low concentrations in one sample, but pyrethroids were below detection limits. Interestingly, while no pyrethroids were detected in the water sample collected from site Cache-Ulatis on Jan. 31, 2008 and stored at ATL for the duration of the initial toxicity test, analysis of a sample from this site collected for CVRWQCB at the same time, and sent for chemical analysis the following day, contained two pyrethroids, 0.007 µg/L cyfluthrin and 0.003 µg/L bifenthrin, and one organophosphate insecticide, 0.011 µg/L disulfoton. This demonstrates that degradation of pesticides takes place even if water samples are preserved with DCM, and stored in the dark at 4^{o}C until toxicity test results become available. This poses problems for the identification/confirmation of toxicity testing results by analytical chemistry, especially for low or sublethal (growth) levels of toxicity.

III.2.2.3 Chronic Toxicity to *H. azteca* - Effects on 10-d Growth

Chronic toxicity in the form of reduced amphipod growth compared to control was detected in 7 samples (0.9 % of total) without apparent seasonal or geographic patterns (Table 6 a, b). Amphipods exposed to a sample from Cache-Ulatis (collected Jan. 31, 2008) not only weighed significantly less than control animals, but PBO addition also caused significantly higher mortality than the ambient sample alone. This suggested that pyrethroid insecticides were present at concentrations that caused sublethal toxicity. In general, however, the growth endpoint is not a sensitive indicator of toxicity due to the variable size of the organisms, the variability of food content in Delta water samples, and the lack of natural particulate matter in the laboratory control water.

PBO Effect on 10-d Growth: A total of 100 samples (13.3 %) demonstrated significant PBO effects on amphipod growth, 45 in 2008 and 55 in 2009. Of these, 36 (4.8 % of total samples tested) showed an increase in weight with PBO addition (antagonistic PBO effect), and 64 (8.5 % of total) a decrease (synergistic PBO effect). Most samples resulting in a significant reduction or increase in growth were submitted for chemical analysis.

Table 5. List of samples causing significant reduction in 10-d survival of *H. azteca* or PBO effects on survival.

Site	Sampling Date	10-d Survival ± SE (%)						
		Control	EC Control	(EC) Control w. PBO	Ambient	Ambient w. PBO (% Ambient)	TIE Results	Analytical Results
340	May 13 2009	100±0.0	73±6.0	73±11.1	61±10.1	14±9.0 [a, b]	N/A	**P:** ND
405	Mar 27 2008	100±0.0	100±0.0	93±4.8	87±2.2 [a]	97±2.8	N/A	N/A
602	July 20 2009	100±0.0	97±2.8	100±0.0	93±4.8	82±4.3 [a]	N/A	**O + P:** ND
"	Sept 16 2009	91±3.0	98±2.5	100±0.0	95±2.9	87±2.4 [a]	N/A	N/A
711	June 25 2009	97±3.1	89±6.4	77±6.1	45±7.6 [a]	61±4.2 [a]	N/A	**C:** Diuron: 3 ng/L
"	July 9 2009	98±2.5	100±0.0	98±2.5	93±3.3	83±2.2 [a]	N/A	N/A
902	Aug 28 2008	84±3.1	N/A	94±6.3	97±3.1	28±20.7 [a, b]	N/A	**P:** Cypermethrin: 16 ng/L
"	June 25 2009	97±3.1	N/A	95±2.8	90±7.1	85±4.2 [a]	N/A	Not analyzed.
915	Aug 28 2008	84±3.1	N/A	94±6.3	94±3.6	60±14.1 [a, b]	N/A	**P:** ND
CU	Jan 31 2008	95±2.8	N/A	95±2.9	97±2.8	8±4.9 [a, b]	PBO Addition Increased Toxicity @ 15°	ND*
"	Feb 28 2008	92±4.8	N/A	98±2.5	98±2.5	66±9.3 [a, b]	N/A	**P:** Bifenthrin: 1 ng/L, Lambda-cyhalothrin: 1 ng/L
"	Oct 15 2009	93±7.5	N/A	92±4.8	98±2.5	10±6.1 [a, b]	C8 Eluate Toxic, C8 Rinsate Reduced Toxicity, PBO Addition Increased Toxicity, More Toxicity at 15° than 23°	**O + P:** Chlorpyrifos: 3 ng/L, Diazinon: 3 ng/L
Hood	June 23 2009	92±2.7	95±2.6	91±5.1	87±3.0	66±6.1 [a, b]	N/A	**P:** Permethrin: 20 ng/L (7 cis, 13 trans)
Napa	Jun 17 2008	97±2.8	95±2.9	98±2.5	89±6.8	82±2.7 [a]	N/A	**C:** Diuron: 32 ng/L, **M**: See Below
Suisun @ Rush R.	Aug 12 2008	98±2.5	98±2.5	95±2.9	16±6.5 [a]	0±0.0 [a]	C8 Eluate Toxic; PBO Addition Increased Eluate Toxicity	**P**: ND **M**: See Below
Light 55	Aug 14 2008	100±0.0	N/A	100±0.0	85±8.7 [a]	97±3.3	N/A	Not analyzed.

[a] Significantly different from appropriate control ($p \leq 0.05$);

[b] Significantly different from ambient sample ($p \leq 0.05$);

Chemical Analysis Key: P: Pyrethroid, O: Organophosphate, C: Comprehensive, Cb: Carbamate, M: Metal

*Analysis of a water sample (CVRWQCB) without storage resulted in the detection of 0.007 µg/L cyfluthrin, 0.003 µg/L bifenthrin & 0.011 µg/L disulfoton

Table 6 a. Samples collected in 2008 and showing significant differences in final *H. azteca* weight compared to controls or PBO effects, and corresponding analytical results. Gray shade: significant difference from control; pink shade: significant increase in weight due to PBO.

Site	Sampling Date	10-d Weight ± SE (µg/individual)					
		Test Acceptability Control	EC Control	(EC) Control with PBO	Ambient	Ambient with PBO	Analytical Results
340	Mar 27 2008	46±8	37±1	42±12	47±1	66±2 [b]	N/A
"	Apr 10 2008	42±6	24±14	43±9	50±7	21±3 [b]	N/A
405	Feb 13 2008	44±6	76±17	85±5	69±5	46±3 [b]	N/A
"	May 23 2008	37±14	27±12	48±7	57±8	24±6 [b]	P: ND
508	Mar 27 2008	46±8	N/A	55±4	100±5	77±7 [b]	N/A
"	May 7 2008	42±6	N/A	66±3	103±4	71±6 [b]	P: ND
"	May 23 2008	37±14	N/A	25±10	66±8	91±2 [b]	N/A
"	June 4 2008	53±3	49±4	45±3	101±8	78±4 [b]	N/A
"	July 3 2008	118±16	N/A	66±11	68±8 [a]	76±22	N/A
"	Aug 27 2008	46±2	N/A	43±5	63±3	49±4 [b]	N/A
"	Sept 10 2008	46±6	N/A	73±9	91±10	57±5 [b]	P: ND
"	Oct 8 2008	80±12	N/A	53±5	47±8 [a]	48±5	N/A
602	May 7 2008	42±6	81±9	66±3	62±4	44±3 [b]	N/A
"	May 23 2008	37±14	40±5	54±14	33±10	61±4 [b]	N/A
"	June 4 2008	53±3	49±4	45±3	69±4	48±7 [b]	N/A
609	Apr 24 2008	17±8	N/A	25±3	70±4	43±3 [b]	N/A
"	July 16 2008	53±6	81±7	47±10	93±7	47±8 [b]	P: ND
"	July 30 2008	66±8	89±12	68±7	106±10	66±11 [b]	P: ND
"	Sept 10 2008	46±6	46±2	50±8	72±5	36±9 [b]	P: ND
"	Oct 8 2008	80±12	N/A	53±5	43±12 [a]	22±5 [a]	Cb: 8 ng/L Diuron; Metals: See Below
711	July 3 2008	118±16	105±17	98±3	80±6	28±5 [a, b]	N/A
"	Nov 20 2008	47±13	77±10	47±6	77±10	47±6 [b]	P: ND
815	May 12 2008	44±5	N/A	34±9	71±3	88±4 [b]	N/A
"	May 23 2008	37±14	N/A	25±10	76±3	110±4 [b]	N/A
"	July 17 2008	31±9	35±9	34±6	85±11	36±7 [b]	P: ND
902	May 23 2008	37±14	N/A	25±10	53±16	98±5 [b]	O: ND
915	Jan 2 2008	32±15	N/A	47±9	81±5	100±4 [b]	N/A
"	June 19 2008	73±4	N/A	60±5	157±10	113±11 [b]	N/A
"	July 31 2008	38±5	N/A	45±6	89±9	58±7 [b]	P: ND
"	Aug 28 2008	17±7	N/A	28±7	89±8	42±11 [b]	P: ND
CL	Dec 4 2008	41±8	N/A	35±6	65±5	47±5 [b]	N/A
CU	Jan 31 2008	60±3	N/A	45±10	42±7 [a]	-	ND
"	Mar 13 2008	37±3	N/A	41±8	78±5	48±3 [b]	N/A
"	Mar 26 2008	53±2	N/A	46±5	108±8	77±7 [b]	N/A
"	Apr 9 2008	35±3	N/A	31±5	56±4	34±4 [b]	N/A
Hood	Jan 1 2008	32±15	N/A	47±9	34±9	62±6 [b]	N/A
"	July 15 2008	53±6	75±6	53±8	89±6	48±3 [b]	P: ND
"	Oct 7 2008	80±12	41±9	59±9	22±5	50±6 [b]	O: ND
"	Nov 4 2008	41±6	51±11	93±66	91±6	35±10 [b]	P: 1.2 ng/L Cyfluthrin

Table 6 a, cont. Samples collected in 2008 and showing significant differences in final *H. azteca* weight compared to controls or PBO effects, and corresponding analytical results.

Site	Sampling Date	10-d Weight ± SE (µg/individual)					
		Test Acceptability Control	EC Control	(EC) Control with PBO	Ambient	Ambient with PBO	Analytical Results
Light 55	Feb 14 2008	53±6	N/A	56±7	94±3	70±4 [b]	N/A
"	Mar 13 2008	37±3	N/A	41±8	75±7	53±4 [b]	N/A
"	July 17 2008	31±9	35±9	34±6	42±4	62±4 [b]	N/A
Napa	Apr 23 2008	29±5	N/A	27±9	46±8	82±5 [b]	N/A
"	May 22 2008	84±7	63±17	85±6	116±7	55±4 [a]	P: ND
"	July 15 2008	53±6	44±3	69±14	34±3 [a]	43±12	P: ND
R&R	Aug 26 2008	46±2	N/A	43±5	83±3	63±5 [b]	N/A
"	Sept 23 2008	64±4	N/A	63±5	132±16	84±10 [b]	N/A
"	Nov 4 2008	41±6	N/A	35±2	91±8	41±11 [b]	P: ND
Suisun Rush R.	Apr 23 2008	29±5	N/A	27±9	42±4	77±2 [b]	N/A
"	Aug 26 2008	46±2	51±9	38±5	54±5	37±2 [b]	N/A
Suisun Pub. Dock	Jan 15 2008	55±4	N/A	53±4	88±6	104±4 [b]	N/A

[a] Significantly different from appropriate control ($p \leq 0.05$);

[b] Significantly different from ambient sample ($p \leq 0.05$);

Chemical Analysis: P: Pyrethroid, O: Organophosphate, C: Comprehensive, Cb: Carbamate, M: Metal

Table 6 b. Samples collected in 2009 and showing significant differences in final *H. azteca* weight compared to controls or PBO effects, and corresponding analytical results. Gray shade: significant difference from control; pink shade: significant increase in weight due to PBO.

Site	Sampling Date	10-d Weight ± SE (µg/individual)					
		Test Acceptability Control	EC Control	(EC) Control with PBO	Ambient	Ambient with PBO	Analytical Results
340	Feb 18 2009	39±4	25±2	41±21	23±4	52±7 [b]	O: ND
"	Mar 17 2009	48±9	N/A	63±11	73±2	57±5 [b]	N/A
"	Apr 29 2009	55±9	20±5	34±12	48±7	70±5 [b]	N/A
405	Mar 17 2009	48±9	N/A	63±11	75±5	61±3 [b]	N/A
"	Apr 14 2009	46±3	N/A	33±5	30±7 [a]	44±8	N/A
"	July 20 2009	72±8	28±8	38±3	31±6	66±7 [b]	N/A
508	Jan 21 2009	45±6	54±3	40±5	45±6	73±4 [b]	O: ND
"	Feb 5 2009	46±4	29±6	34±4	28±4	46±3 [b]	N/A
"	Mar 4 2009	57±7	N/A	46±5	131±6	83±13 [b]	P: ND
602	Apr 29 2009	55±9	N/A	69±6	54±7	81±4 [b]	O: ND
"	Apr 20 2009	72±8	58±4	43±5	46±1 [a]	74±5 [b]	O+P: ND
"	Nov 10 2009	46±5	35±4	31±3	35±5	55±6 [b]	O+P: ND
609	May 27 2009	35±6	N/A	78±6	90±1	75±5 [b]	N/A
711	Jan 22 2009	75±6	N/A	53±9	107±6	78±8 [b]	P: ND
"	Sept 15 2009	43±8	42±7	33±3	100±5	75±9 [b]	O+P: ND
815	Feb 19 2009	34±6	N/A	30±6	56±5	98±7 [b]	O: Diazinon: 2 ng/L
"	Aug 20 2009	30±7	N/A	38±2	112±3	65±18 [b]	N/A
902	Jan 22 2009	75±6	N/A	53±9	127±6	75±8 [b]	P: ND
"	Feb 4 2009	66±9	N/A	52±3	119±14	44±12 [b]	P: ND
"	Apr 2 2009	50±8	N/A	50±12	90±7	124±7 [b]	O: Chlorpyrifos: 2 ng/L (Below RL); Disulfoton: 8 ng/L (Below RL)
"	June 11 2009	27±9	29±8	42±7	81±4	60±9 [b]	P: Cypermethrin: 2 ng/L
"	Oct 15 2009	49±9	N/A	32±7	88±7	49±6 [b]	O+P: ND
915	Jan 22 2009	75±6	N/A	53±9	84±16	127±5 [b]	O: ND
"	Mar 18 2009	26±5	N/A	46±5	93±6	69±4 [b]	P: ND
"	June 25 2009	46±10	N/A	44±5	55±10	78±7 [b]	O: ND
"	Oct 27 2009	55±8	N/A	45±7	61±5	116±11 [b]	O+P: 1.5 ng/L Lambda-cyhalothrin; Permethrin: 48.9 ng/L (15.1 cis, 33.8 trans)
CL	Feb 4 2009	66±9	N/A	52±3	105±4	60±2 [b]	P: ND
"	June 25 2009	46±10	N/A	44±5	83±7	43±7 [b]	P: ND
"	Aug 20 2009	30±7	26±5	32±3	101±8	72±8 [b]	N/A
"	Oct 15 2009	38±4	N/A	44±2	83±5	62±4 [b]	O+P: ND
"	Dec 30 2009	51±4	N/A	88±16	96±10	127±8 [b]	O: ND
CU	Feb 4 2009	66±9	N/A	52±3	121±11	63±5 [b]	P: ND
"	Feb 19 2009	34±6	N/A	30±6	74±8	42±6 [b]	P: Bifenthrin: 117 ng/L
"	Mar 5 2009	45±2	N/A	60±26	73±4	40±5 [b]	P: ND
"	Apr 2 2009	50±8	N/A	50±12	36±5	106±5 [b]	O: Chlorpyrifos: 78 ng/L; Disulfoton: 17 ng/L (Below RL)
Hood	Mar 19 2009	26±5	N/A	46±5	92±3	67±6 [b]	P: Permethrin: 3 ng/L (2 cis, 1 trans)

Table 6 b, cont. Samples collected in 2009 and showing significant differences in final *H. azteca* weight compared to controls or PBO effects, and corresponding analytical results. Gray shade: significant difference from control; pink shade: significant increase in weight due to PBO.

Site	Sampling Date	10-d Weight ± SE (µg/individual)					
		Test Acceptability Control	EC Control	(EC) Control with PBO	Ambient	Ambient with PBO	Analytical Results
"	Apr 28 2009	55±9	55±2	57±6	77±5	99±5 [b]	O: ND
"	July 7 2009	67±4	57±6	58±4	94±5	56±6 [b]	P: Permethrin: 11 ng/L (5 cis, 6 trans)
"	Sept 17 2009	60±11	59±4	76±10	104±2	81±3 [b]	O+P: ND
"	Oct 1 2009	42±12	N/A	52±4	51±9	77±4 [b]	O+P: ND
"	Dec 3 2009	51±5	N/A	50±6	101±4	84±7 [b]	O+P: ND
Light 55	Feb 4 2009	66±9	N/A	52±3	79±5	50±5 [b]	P: ND
"	Mar 18 2009	48±9	N/A	63±11	72±4	97±3 [b]	O: Chlorpyrifos: 10 ng/L
"	June 11 2009	27±9	N/A	37±3	86±6	64±2 [b]	P: Cypermethrin: 2 ng/L
"	Dec 1 2009	37±5	N/A	41±3	74±6	99±7 [b]	O+P: 1.4 ng/L Esfenvalerate
"	Dec 15 2009	34±3	N/A	30±5	68±4	43±5 [b]	P: ND
NAPA	July 7 2009	67±4	31±1	33±5	39±4	12±2 [a, b]	N/A
R&R	Jan 6 2009	42±6	N/A	76±9	117±6	64±15 [b]	P: ND
"	Mar 17 2009	48±9	N/A	63±11	93±6	64±7 [b]	P: Cyfluthrin: 3 ng/L
"	Aug 18 2009	45±5	36±6	27±6	100±12	66±7 [b]	P: Cyfluthrin: 0.4 ng/L
"	Sept 17 2009	60±11	N/A	70±6	135±4	117±3 [b]	N/A
Suisun @ Rush R.	Feb 17 2009	39±4	N/A	32±6	35±6	60±1 [b]	O: Disulfoton: 14 ng/L
"	Apr 15 2009	65±6	N/A	56±1	50±14	90±5 [b]	O: ND
"	Apr 28 2009	55±9	N/A	69±6	90±6	119±6 [b]	O: ND
"	Dec 3 2009	51±5	43±3	40±5	124±12	78±5 [b]	O+P: ND
"	Dec 17 2009	45±6	N/A	37±8	62±2	76±1 [b]	O: ND

[a] Significantly different from appropriate control ($p \leq 0.05$);

[b] Significantly different from ambient sample ($p \leq 0.05$);

Chemical Analysis: P: Pyrethroid, O: Organophosphate, C: Comprehensive, Cb: Carbamate, M: Metal

Site-specific toxicity: Information on the number of samples causing significant effects on amphipod survival and growth, or PBO effects at any given sampling site is summarized in Table 7. Cache-Ulatis had the greatest number of samples, where a PBO effect on survival suggested the presence of pyrethroid insecticides. In addition, PBO synergized effects on growth in 6 samples. Sites Hood, Cache-Ulatis, Rough and Ready Island, Suisun Slough at Rush Ranch, Light 55, 915 and 508 had the greatest number of samples with synergistic or antagonistic effects on amphipod growth.

Table 7. Number of samples causing significant differences in *H. azteca* survival or growth by sampling site.

Site	Total Samples Tested *[n]*	Reduced Survival[1] *[n]* (% of Total)	Reduced Survival (PBO)[2] *[n]*	PBO Effect on Survival *[n]*	Reduced Weight[1] *[n]* (% of Total)	Synergistic PBO Effect on Growth *[n]*	Antagonistic PBO Effect on Growth *[n]*
340	17	0	1	1 (6%) ↓	0	2	3
405	32	1 (3%)	0	0	1 (3%)	3	1
508	51	0	0	0	2 (4%)	6	3
602	51	0	2	0	1 (2%)	2	4
609	50	0	0	0	1 (2%)	5	0
711	51	1 (2%)	2	0	0	4	0
815	50	0	0	0	0	2	3
902	50	0	2	1 (2%) ↓	0	4	2
910	1	0	0	0	0	0	0
915	50	0	1	1 (2%) ↓	0	4	4
CL	51	0	0	0	0	5	1
CU	51	0	3	3 (6%) ↓	1 (2%)	6	1
Hood	51	0	1	1 (2%) ↓	0	6	4
Light 55	51	1 (2%)	0	0	0	5	3
Napa	36	0	1 (3%)	0	1 (2.8%)	1	1
R & R	50	0	0	0	0	7	0
Suisun @ Rush R.	49	1 (2%)	0	*	0	2	5
Suisun Pub. Dock	2	0	0	0	0	0	1

[1] Ambient samples significantly different from appropriate control.
[2] Ambient samples with PBO significantly different from PBO control.
↓ Synergistic effect of PBO
↑ Antagonistic effect of PBO

Effect of ammonia/um on amphipod survival and growth: Correlation analysis of data collected over a 4-year period (2006-2010) revealed significant relationships between amphipod survival and growth at several sites (Table 8). Total ammonia/um concentrations were positively correlated with amphipod survival at sites 323, 504, 804, 915 and Cache-Lindsey, but negatively correlated at Cache-Ulatis. Amphipod growth was negatively correlated with total ammonia/um at sites 405, 609, 711, Light 55, Napa and Rough and Ready Island, and positively correlated at site 910. Un-ionized ammonia was negatively correlated with amphipod growth at Rough and Ready Island.

Table 8. Site-specific correlation between total and un-ionized ammonia and *H. azteca* survival and growth in Delta water samples collected 2006 - 2009.

Site	Survival vs Total NH_4^+/NH_3		Survival vs NH_3		Log_{10} Weight vs NH_4^+/NH_3		Log_{10} Weight vs NH_3	
	Coeff.	*P*	Coeff.	*P*	Coeff.	*P*	Coeff.	*P*
323	0.726	0.017*	0.558	0.094	-0.038	0.861	3.114	0.775
340	-0.181	0.346	0.061	0.760	-0.001	0.983	-4.333	0.484
405	-0.339	0.078	-0.295	0.128	-0.057	0.032*	-3.051	0.171
504	0.578	<0.0001*	0.219	0.149	0.032	0.678	0.410	0.929
508	0.059	0.559	-0.034	0.737	-0.026	0.407	-2.959	0.153
602	0.183	0.177	0.025	0.854	-0.065	0.050	-1.760	0.454
609	0.204	0.052	0.118	0.266	-0.052	0.039*	-4.029	0.181
704	0.242	0.091	0.005	0.974	0.037	0.538	0.641	0.858
711	0.095	0.348	0.097	0.338	-0.036	0.049*	-0.881	0.131
804	0.354	0.012*	0.010	0.943	0.020	0.761	-2.805	0.262
812	0.166	0.260	0.043	0.771	-0.108	0.158	-3.088	0.361
815	0.060	0.676	-0.009	0.950	-0.006	0.896	-0.036	0.984
902	0.012	0.905	-0.110	0.276	-0.044	0.250	2.546	0.136
910	0.207	0.150	-0.154	0.285	0.099	0.029*	1.163	0.685
915	0.219	0.029*	0.075	0.460	0.019	0.609	1.745	0.492
CL	0.340	0.014*	0.089	0.529	-0.020	0.476	-1.210	0.087
CU	-0.131	0.350	-0.283	0.040*	-0.085	0.089	0.159	0.926
Hood	0.056	0.674	-0.016	0.907	0.019	0.408	-0.781	0.446
Light 55	-0.168	0.098	-0.164	0.107	-0.068	0.018*	-1.191	0.226
Napa	-0.210	0.375	-0.306	0.190	-0.070	0.021*	-2.909	0.525
R&R	-0.159	0.271	0.098	0.501	-0.099	0.011*	-6.301	0.012*
Suisun @ Rush R.	0.072	0.652	0.089	0.576	-0.008	0.824	-4.145	0.268

Survival: Nonparametric correlations in the EC < 12,000 range.
Weight: Multivariate regression models with EC controlled as a covariate with linear and quadratic terms.

III.2.2.4 Analytical Chemistry

When acute toxicity was observed, whole water samples preserved with DCM were immediately submitted to CDFG-WPCL for chemical analysis. Otherwise, samples were

submitted after statistical analysis was complete and significant PBO effects on growth were detected (approx. 14 d after sample collection). Ambient water samples submitted for chemical analysis are listed in Tables 9 a-c. For a detailed list of analytes, please see Table A1, Appendix A. A total of 113 samles were submitted for analysis. Pyrethroids, in particular cyfluthrin, permethrin, cypermethrin, bifenthrin, lambda-cyhalothrin and esfenvalerate, were detected in 24 samples, organophosphates, in particular chlorpyrifos, diazinon and disulfoton, were detected in 13 samples, and the herbicide diuron was detected in all ten samples it was analyzed in. Numerous samples contained more than one contaminant.

Pyrethroids were not detected in several samples, where TIE analyses strongly suggested that toxicity was due to this group of insecticides. Pyrethroids and organophosphates were, however, detected in some of the samples that showed reductions or increases in *H. azteca* weight, respectively. Pyrethroid insecticides were detected in low concentrations from samples collected at Rough & Ready Island on 3/17/2009 (3 ng/L cyfluthrin) and Hood on 3/18/2009 (3 ng/L permethrin). The organophosphate insecticides chlorpyrifos, diazinon, and disulfoton were detected singularly or in combination at sites 508, 602, 815, 902, Cache-Ulatis, and Light 55. Although the majority of these detections were below the reporting limit of the analytical laboratory, a sample collected from Cache-Ulatis on 4/2/2009 resulted in the detection of 78 ng/L chlorpyrifos. This sample was submitted to CDFG-WPCL following a significant increase in growth with the addition of PBO in the 10-d *H. azteca* bioassay. The detected concentration of chlorpyrifos is greater than the 10-d control water LC50 of 67.2 ng/L determined by UCD-ATL in January 2009, but survival was not affected in this test. A sample collected from Light 55 on 3/19/09 caused a significant increase in growth when treated with PBO and resulted in the detection of 10 ng/L chlorpyrifos. The herbicide diuron was detected at concentrations of up to 32 ng/L (Napa - Aug. 12, 2008). Analytical results of total and dissolved metals suggest that metals are not the dominant toxicants in these water samples, although a more detailed analysis of available LC50 and effect concentrations at different salinities, as well as mixture effects is needed.

In many samples where survival or weight effects suggested the influence of insecticides, chemical analyses resulted in no detection of chemicals. This may, in part, be due to the very high sensitivity of *H. azteca* to pyrethroid insecticides. For example, effect concentrations of bifenthrin and cyfluthrin are close to the reporting and detection limits of the chemical analysis (Table 10). Analyte degradation may have further reduced our capability to detect the small amounts of pesticide capable of affecting *H. azteca*. Although samples destined for pyrethroid analysis were preserved with DCM within 12 hours of collection, the time from sample collection to observation of toxicity caused a latency of approximately two weeks from sample collection to delivery to the analytical laboratory.

Table 9 a. Results of chemical analysis of whole water samples analyzed in 2008 and 2009. Samples are listed in chronological order. All samples were submitted approx. 14 d after collection unless otherwise noted. Results of metals analysis provided in Table 4-5 b. ND=below detection limit; * detection below reporting limit

Site	Collection Date	Scan Type	Results
CU	1/31/2008	pyrethroid scan	ND
CU	1/31/2008	OP & Pyrethroids	7 pptr cyfluthrin, 3 pptr bifenthrin, 11 pptr disulfoton (submitted within 24 h of sample collection)
CU	2/28/2008	pyrethroid scan	1 pptr bifenthrin, 1 pptr lambda-cyhalothrin
711	4/9/2008	comprehensive[1]	1 pptr bifenthrin, 41 pptr diuron + METALS
CL	4/9/2008	comprehensive	86 pptr diuron, + METALS
Hood	4/22/2008	comprehensive	PAH: 0.0473 ng/mL naphthalene, 0.0141 ng/mL methylnaphthalene, 2-, 0.00769 ng/mL methylnaphthalene, 1-, 0.0232 ng/mL naphthalenes, C1-, 0.0141 naphthalenes, C2- 1 pptr lambda-cyhalothrin, 87 pptr diuron, + METALS PAH: 0.005 ng/mL fluoranthene, 0.008 ng/mL benzo(b)fluoranthene
CL	4/23/2008	comprehensive	1 pptr bifenthrin, 60 pptr diuron, 4 pptr methomyl, + METALS PAH: 0.011 ng/mL fluoranthene, 0.010 ng/mL fluoranthene/pyrenes, C1-, 0.009 ng/mL pyrene, 0.007 ng/mL benz(a)anthracene, 0.007 ng/mL chrysene, 0.017 ng/mL benzo(b)fluoranthene, 0.010 ng/mL benzo(e)pyrene, 0.010 ng/mL benzo(a)pyrene, 0.012 ng/mL indenol(1,2,3-c,d)pyrene, 0.012 ng/mLbenzo(g,h,i)perylene
711	4/23/2008	comprehensive	1 pptr bifenthrin, 60 pptr diuron, 3 pptr carbaryl + METALS
508	5/7/2008	pyrethroid scan	ND
902	5/12/2008	comprehensive	57 pptr diuron + METALS
Napa	5/21/2008	pyrethroid scan	ND
405	5/23/2008	pyrethroid scan	ND
902	5/23/2008	OP scan	ND
Napa	6/17/2008	comprehensive	32 pptr diuron + METALS
Napa	7/15/2008	pyrethroid scan	ND
Hood	7/15/2008	pyrethroid scan	ND
609	7/16/2008	pyrethroid scan	ND
815	7/17/2008	pyrethroid scan	ND
405	7/30/2008	comprehensive[1]	10 pptr diuron + METALS
609	7/30/2008	pyrethroid scan	ND
915	7/31/2008	pyrethroid scan	ND
902	7/31/2008	pyrethroid scan	ND
Suisun	8/12/2008	pyrethroid scan & metals	ND + METALS
602	8/13/2008	pyrethroid scan	ND
902	8/28/2008	pyrethroid scan	16 pptr cypermethrin
915	8/28/2008	pyrethroid scan	ND
508	9/10/2008	pyrethroid scan	ND
609	9/10/2008	pyrethroid scan	ND
609	9/24/2008	pyrethroid scan	ND

Site ID	Collection Date	Scan Type	Results
Hood	10/7/2008	organophosphate scan	ND
609	10/8/2008	carbamate scan & metals	8 pptr diuron + METALS
Hood	11/4/2008	pyrethroid scan	1.2 pptr cyfluthrin
R&R	11/4/2008	pyrethroid scan	ND
711	11/20/2008	pyrethroid scan	ND
R&R	1/6/2009	pyrethroid	ND[2]
602	1/7/2009	organophosphate	8 pptr disulfoton*
508	1/21/2009	organophosphate	ND
711	1/22/2009	pyrethroid	ND
915	1/22/2009	organophosphate	ND
902	1/22/2009	pyrethroid	ND
Hood	1/23/2009	pyrethroid	ND
Cache-Ulatis	2/4/2009	pyrethroid	ND
902	2/4/2009	pyrethroid	ND
Cache-Lindsey	2/4/2009	pyrethroid	ND
Light 55	2/4/2009	pyrethroid	ND
Suisun	2/17/2009	organophosphate	14 pptr disulfoton
340	2/18/2009	organophosphate	ND
815	2/19/2009	organophosphate	2 pptr diazinon
Cache-Ulatis	2/19/2009	pyrethroid	117 pptr bifenthrin
508	3/4/2009	pyrethroid	ND
Cache-Ulatis	3/5/2009	pyrethroid	ND
Rough & Ready	3/17/2009	pyrethroid	3 pptr cyfluthrin
815	3/18/2009	organophosphate	2 pptr diazinon*, 3 pptr chlorpyrifos*, 8 pptr disulfoton*
Hood	3/18/2009	pyrethroid	3 pptr permethrin
915	3/18/2009	pyrethroid	ND
Light 55	3/19/2009	organophosphate	10 pptr chlorpyrifos
508	4/1/2009	organophosphate	2 pptr chlorpyrifos*
902	4/2/2009	organophosphate	2 pptr chlorpyrifos*, 8 pptr disulfoton*
Cache-Ulatis	4/2/2009	organophosphate	78 pptr chlorpyrifos, 17 pptr disulfoton*
Suisun	4/15/2009	organophosphate	ND
Suisun	4/28/2009	organophosphate	ND
Hood	4/28/2009	organophosphate	ND
602	4/29/2009	organophosphate	ND
340	5/13/2009	pyrethroid	ND
Napa	6/9/2009	pyrethroid	9 pptr esfenvalerate/fenvalerate
340	6/10/2009	organophosphate	ND
Light 55	6/11/2009	pyrethroid	2 pptr cypermethrin
902	6/11/2009	pyrethroid	2 pptr cypermethrin

Site ID	Collection Date	Scan Type	Results
Hood	6/23/2009	pyrethroid	20 pptr permethrin (7 pptr cis, 13 pptr trans)
R&R	6/23/2009	organophosphate	12 pptr diazinon
711	6/25/2009	comprehensive[1]	3 pptr diuron + METALS results not yet available
Cache-Lindsay	6/25/2009	pyrethroid	ND
902	6/25/2009	METALS	results not yet available
915	6/25/2009	organophosphate	ND
Hood	7/7/2009	pyrethroid	11 pptr permethrin (5 pptr cis, 6 pptr trans)
602	7/20/2009	OP + pyrethroid	ND
711	8/6/2009	METALS	results not yet available
R&R	8/18/2009	pyrethroid	0.4 pptr cyfluthrin
CL	8/20/2009	pyrethroid	2 pptr cypermethrin
815	8/20/2009	pyrethroid	2 pptr cypermethrin
CL	9/1/2009	pyrethroid	ND
CU	9/1/2009	pyrethroid	ND
711	9/15/2009	pyrethroid	ND
Hood	9/17/2009	pyrethroid	ND
609	9/16/2009	OP + pyrethroid	ND[1]
CL	9/15/2009	OP + pyrethroid	ND[1]
609	9/30/2009	OP + pyrethroid	2.6 pptr bifenthrin, 1.4 pptr cypermethrin
R&R	10/1/2009	OP + pyrethroid	1.7 pptr bifenthrin
Hood	10/1/2009	OP + pyrethroid	ND
CU	10/15/2009	OP + pyrethroid	3 pptr diazinon, 3 pptr chlorpyrifos
CL	10/15/2009	OP + pyrethroid	ND
902	10/15/2009	OP + pyrethroid	ND
815	10/15/2009	OP + pyrethroid	2 pptr chlorpyrifos, 5 pptr diazinon
Light 55	10/15/2009	OP + pyrethroid	ND
Suisun	10/16/2009	OP + pyrethroid	ND
R&R	10/16/2009	OP + pyrethroid	ND
CU	10/27/2009	OP + pyrethroid	34.7 pptr permethrin (16.5 cis, 18.2 trans), 2 pptr chlorpyrifos
915	10/27/2009	OP + pyrethroid	1.5 pptr lambda-cyhalothrin, 48.9 pptr permethrin (15.1 cis, 33.8 trans)
CL	10/27/2009	OP + pyrethroid	ND
R&R	10/29/2009	OP + pyrethroid	ND
Hood	10/29/2009	OP + pyrethroid	3.7 pptr permethrin (3.7 trans)
CL	11/9/2009	OP + pyrethroid	ND
815	11/9/2009	OP + pyrethroid	ND
602	11/10/2009	OP + pyrethroid	ND
711	12/1/2009	OP + pyrethroid	ND
Light 55	12/1/2009	OP + pyrethroid	1.4 pptr esfenvalerate
Suisun	12/3/2009	OP + pyrethroid	ND
Hood	12/3/2009	OP + pyrethroid	ND
Light 55	12/17/2009	pyrethroid	ND
Suisun	12/17/2009	pyrethroid	ND
CL	12/30/2009	pyrethroid	ND

[1] delivered to DFG on furlough Friday, samples were not refrigerated for 2 d and extraction delayed.

Table 9 b. Results of metals analysis of toxic samples collected 2008 - 2009.

Metal		711	CL	Hood	CL	711	902	Napa	405	Suisun (RR)	609
		4/9/2008	4/9/2008	4/22/208	4/23/2008	4/23/2008	5/12/2008	6/17/2008	7/30/2008	8/12/2008	10/8/2008
Ag	Total	<0.001	0.002	<0.001	<0.001	0.005	<0.001	0.12	0.08	0.67	<0.02
	Dissolved	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	0.48	0.09	0.04	<0.02
Al	Total	206	771	307	1421	398	613	1970	1431	15256	536
	Dissolved	5.89	**11.3**	**13.5**	<1.7	2.71	**20.5**	15.1	<1.7	<1.7	<1.7
As	Total	2.03	2.6	1.84	2.46	1.93	2.69	55.1	66.4	39.5	27.3
	Dissolved	2.58	2	1.77	2.42	1.98	2.24	49.8	63.7	40.3	25.6
Cd	Total	0.025	0.035	0.02	0.017	0.016	0.02	0.32	0.26	0.49	0.11
	Dissolved	0.014	0.016	0.01	<0.004	<0.004	0.01	0.32	0.19	0.29	0.10
Cr	Total	2.46	6.11	3.1	4.25	1.62	2.91	11.8	9.13	27.1	4.51
	Dissolved	**3.6**	0.64	0.86	1.22	0.85	1.28	5.06	5.57	3.70	3.25
Cu	Total	3.01	4.1	2.22	4.41	2.16	3.47	57.9	63.3	61.6	24.9
	Dissolved	2.4	1.51	1.25	2.13	1.3	1.72	49.0	58.9	37.2	21.2
Mn	Total	26.3	37.6	34.5	43.3	24.9	26.7	557	49.4	1231	89.1
	Dissolved	5.29	9.94	6.39	0.19	0.39	0.83	15.0	2.54	34.0	72.0
Ni	Total	2.21	5.41	2.19	6.75	2.22	3.53	22.7	16.2	48.0	8.15
	Dissolved	1.94	1.29	1.02	1.83	1.04	2.13	13.6	11.7	13.4	6.50
Pb	Total	0.31	0.56	0.32	0.62	0.21	1.31	3.13	0.63	20.3	0.41
	Dissolved	**0.18**	0.079	**0.27**	<0.002	0.002	**0.49**	0.11	0.04	<0.015	<0.015
Se	Total	<0.45	<0.45	<0.45	<0.45	<0.45	<0.45	136	150	84.0	44.7
	Dissolved	0.51	0.53	<0.45	0.69	<0.45	0.59	119	137	77.1	55.0
Zn	Total	7.04	6.52	6.37	6.5	2.43	3.91	12.6	5.11	83.4	1.41
	Dissolved	**7.69**	2.08	2.16	0.74	0.57	**5.02**	1.96	2.04	1.00	<0.50

Results still pending for: 711-062509, 902-062509, 711-080609

Table 9 c. Results of routine metals analysis performed during 2009.

Site	Date Collected	Dissolved Metals (ppb)										
		Ag	**Al**	**As**	**Cd**	**Cr**	**Cu**	**Mn**	**Ni**	**Pb**	**Se**	**Zn**
711	2/19/2009	<.02	12.8	1.52	<.01	0.11	2.54	0.79	1.49	0.04	2.24	<.50
711	3/18/2009	0.03	<1.7	2.88	0.14	2.40	2.24	6.57	1.89	<.015	<.60	<.50
711	4/15/2009	<.02	3.28	1.46	<.01	0.18	2.78	0.05	0.85	<.015	<.60	<.50
711	5/28/2009	0.06	2.72	1.60	0.03	<.10	2.23	12.0	0.60	0.03	<.60	<.50
902	2/19/2009	<.02	11.7	2.45	<.01	0.56	2.23	7.64	2.07	0.05	3.68	<.50
902	3/18/2009	<.02	9.88	2.01	0.12	2.07	2.86	7.33	2.67	0.02	<.60	<.50
902	4/23/2009	0.08	11.9	2.16	0.03	<.10	2.69	2.60	1.02	0.14	3.03	<.50
902	5/28/2009	0.05	7.60	2.64	0.05	0.13	3.19	3.25	1.04	0.04	<.60	<.50
CL	2/19/2009	<.02	6.05	2.14	<.01	0.38	2.35	0.27	1.92	0.02	1.86	0.55
CL	3/18/2009	<.02	<1.7	2.58	0.02	2.18	3.35	3.79	2.53	0.02	<.60	<.50
CL	4/15/2009	<.02	5.91	2.25	<.01	<.10	**4.93**	0.13	1.69	0.03	3.01	<.50
CL	5/28/2009	0.07	2.03	1.92	0.05	0.21	2.25	6.68	1.13	0.04	0.89	<.50
Rough & Ready	2/17/2009	<.02	2.68	2.11	<.01	<.10	2.57	0.09	2.64	<.015	3.66	1.21
Rough & Ready	3/17/2009	0.62	<1.7	2.83	0.13	2.17	2.95	0.87	3.28	<.015	1.2	<.50
Rough & Ready	4/14/2009	0.09	2.85	2.85	<.01	<.10	**4.64**	0.27	2.87	0.02	3.51	<.50
Rough & Ready	5/27/2009	0.04	44.7	2.57	0.10	1.28	**4.41**	25.1	2.53	0.03	4.09	<.50
Suisun	2/19/2009	0.04	4.42	10.1	0.02	1.40	11.5	52.1	9.00	0.02	43.6	2.07
Suisun	3/18/2009	<.02	<1.7	12.1	0.10	5.88	9.65	352	9.01	<.015	19.2	<.50
Suisun	4/15/2009	0.50	6.98	7.92	0.01	1.44	18.9	17.4	9.08	0.15	25.3	2.46
Suisun	5/26/2009	0.51	4.77	9.18	0.11	1.25	11.3	83.8	6.23	0.10	28.4	<.50

June 2009 routine metals results still pending

Table 10. Comparison of analytical detection limits and *H. azteca* sensitivities to some organophosphate and pyrethroid pesticides. LC50 / EC25 values were used preferentially, with LOEC substituted when necessary.

Pesticide	Analytical Chemistry		*H. azteca* Toxicity	
	Estimated Detection Limit (ng/L)	Reporting Limit (ng/L)	10-d Survival LC50/LOEC (ng/L)	10-d Weight EC25/LOEC (ng/L)
Chlorpyrifos	2.0	5.0	84.9	> 66
Diazinon	2.0	5.0	2900	2000
Bifenthrin	0.2	0.4	3.3	0.9
Cyfluthrin	0.4	0.8	2.7	1.5
Permethrin	0.6	1.0	59.0	> 80

III.3 Toxicity Monitoring with Delta Smelt (*Hypomesus transpacificus*)

III.3.1 Methods

III.3.1.1 Ambient Toxicity Tests

Test organisms and control water: Tests were performed using larval delta smelt at age ranges of 28-57 d (2008) and 30-55 d (2009). Delta smelt were obtained from the UC Davis Fish Conservation and Culture Laboratory (UCD-FCCL) in Byron, CA. Hatchery water collected from the UCD-FCCL was used for all control treatments. Fish were transported to UCD-ATL following methods described by Werner et al. (2008).

Sampling: Water samples (35 gal/site) were collected as described in Chapter 1 (above) from the DWR water quality monitoring stations at Hood (Sacramento River) and Rough & Ready Island (San Joaquin River), and from sites Light 55, Suisun Slough at Rush Ranch, Napa River at the Vallejo Seawall, and Cache Slough near the confluence with Lindsey Slough (Table 3-1, Figure 3-1). Water collections for delta smelt toxicity testing occurred four times in 2008 (March 25 to May 22, 2008) and six times in 2009 (March 17 to May 28, 2009).

Testing procedures: Test and fish transport protocols followed those developed at UCD-ATL and described in detail by Werner et al. (2008). A flow-through system was used for testing of ambient waters. Upon arrival at UCD-ATL, the transport containers with fish were placed into a temperature-regulated water bath maintained at 16º C. One-liter glass beakers were used to gently move the fish to a bread pan containing hatchery water at a depth of approximately 2 cm. The fish were then carefully removed from the pan using 100 ml beakers and released into the replicate exposure tanks at random, submerging the beaker and allowing the fish to swim freely into the tanks. Twelve fish were placed into each of the tanks containing 7 L of water for a 48-h EC acclimation period. Hatchery water and EC-adjusted hatchery water was used as acclimation and control water. EC was adjusted with distilled water ("Low EC Control") to match the lowest EC of ambient water samples. When turbidity of the hatchery water was below 11 NTUs, Nanno 3600™, a concentrated *Nannochloropsis* algae solution ($68 \cdot 10^9$ cells/ml; Reed Mariculture, Inc. Campbell, CA) was added to increase turbidity in control treatments. Turbidity of the "Low Turbidity Control" was matched daily to the lowest turbidity seen in ambient samples. For tests conducted in 2009, gram-negative and gram-positive antibiotics (Maracyn and Maracyn-2, Virbac AH Inc., Fort Worth TX) were added at the manufacturer's recommended dose throughout the acclimation and testing period. Final concentrations were 5.3 mg/L Maracyn (erythromycin) and 0.26 mg/L Maracyn-2 (minocycline). During acclimation and testing, fish were fed three times per day with 200 µl of *Artemia* and 300 µl of rotifers. At test initiation, water in aquaria was drained to approximately 2 L to allow for an accurate count of living fish. Water quality parameters (EC, pH, temperature, DO, turbidity and ammonia) were measured daily. Dead fish were counted and removed daily. At test termination, surviving fish were counted, euthanized with MS-222, flash-frozen and stored at -80^{o}C for future biomarker analysis.

III.3.1.2 Statistical Analysis

Data from delta smelt tests were analyzed using the USEPA standard single-concentration statistical protocols (US EPA 2002). The US EPA method follows standardized statistical method used in aquatic toxicology monitoring and regulation throughout the United States.

III.3.2 Results

2008 Monitoring Period: Two out of four samples collected at Hood caused significant reductions in delta smelt survival compared to the appropriate controls (Table 11, Appendix C). While control survival was below 60% in two tests (March 25/27, April 22/24), larval delta smelt survival was consistently high in water from Suisun Slough (85-98%) and the Napa River (76-96%). Larval smelt generally did very well in water from the Deep Water Ship Channel at Light 55 (52-92% survival). Good performance at these sites can potentially be explained by two important water quality parameters: salinity and/or high turbidity. Turbidity appears to be the most important factor, since water at Light 55 generally has low conductivity (200-300 µS/cm) albeit not as low as the Sacramento River. However, although Cache Slough had similar conductivity and turbidity conditions as the Deep Water Ship Channel (Chapter III.1), fish survival rates were generally lower in Cache Slough (59-71%), suggesting less favorable water quality. Survival was significantly lower than the appropriate control in two water samples collected from Hood on April 24 and May 22, 2008.

Table 11. Percent survival of *H. transpacificus* after 7-day exposures to Delta water samples. Results marked in bold/shaded boxes are significantly different from their respective control.

Sampling Dates:		**3/25-3/27/08**	**4/8-4/10/08**	**4/22-4/24/08**	**5/20-5/22/08**
Age of Delta Smelt:		**28 days**	**38 days**	**37 days**	**57 days**
Treatment	Mean EC (µS/cm)	Mean 7-day Survival[1]			
Low EC Control	213	48.7	60	37.7	75
Hatchery Rearing Control	909	58.3	72.9	38.3	75
High EC Control	4034	82.6	77.3	60.8	88.8
Low Turbidity Control	3075	56.3	27.1	50	69.2
Sacramento River at Hood DWR Station[2]	185	25	34.5	**20.2**[5]	**46.1**[5]
Deep Water Channel, Light 55[2]	296	91.7	72.9	52.1	77.5
Confluence of Lindsey Sl. And Cache Sl.[2]	282	59.3	67.8	61.9	71.3
Rough and Ready DWR station[2]	536	42.4	47.9	51.3	55
Suisun Slough at Rush Ranch[3]	3928	91.8	87.5	97.7	85
Napa River at Vallejo Seawall (340)[4]	17767	89.7	95.8	76.1	92.5

1. Highlighted areas indicate significant reductions in survival compared to the appropriate EC-specific and turbidity-specific controls.
2. These low conductivity samples were compared to the Low EC Control.
3. Suisun Slough at Rush Ranch was compared to Mid- and High EC Controls, as appropriate.
4. Napa River at Vallejo Seawall was compared to the High EC Control.
5. Hood showed lower survival than both the low EC control and the low turbidity control according to USEPA statistical protocols.

2009 Monitoring Period: With the exception of the first test of the season, which performed poorly, survival of larval delta smelt after 7 d was ≥76.4 in the "low-conductivity controls", ≥70.8 in "mid-conductivity controls", ≥82.5 in "high conductivity controls, and ≥62.5% in "very high conductivity controls" (Table 12; Appendix C). Survival in the "low conductivity/low turbidity control" was generally matched to the water collected from Hood. Delta smelt survival was significantly lower than appropriate conductivity controls in ambient water samples collected from Hood on 4/28/09, from Cache-Lindsey on 4/30/09 and from Rough & Ready collected on 3/31/09, 4/15/09 and 5/12/09. Other instances of significantly reduced survival relative to conductivity-specific controls are samples from Light 55 and Cache-Lindsey, collected on 3/31/09. Water from these sites generally had relatively high turbidity (Chapter III.1). Survival was consistently high in samples collected at the high conductivity, high turbidity site at Suisun Slough at Rush Ranch, as was observed in tests performed in 2008. At site 340, where conductivity was higher and turbidity was lower than in Suisun Slough, survival was generally lower, highlighting the importance of turbidity as a factor influencing delta smelt survival.

We conclude that water quality in the Sacramento River at Hood, in the San Joaquin River at Rough & Ready Island, and near the confluence of Cache and Lindsey Sloughs is at times unfavorable for larval delta smelt. This may in some cases be partly attributable to low turbidity stress. Turbidity, and to a lesser degree salinity/EC, are extremely important parameters influencing larval delta smelt survival. All testing therefore requires controls matching the salinity/EC and turbidity of each sample to obtain conclusive data on water toxicity.

Table 12. Survival of larval *H. transpacificus* in water samples collected from the Sacramento - San Joaquin Delta. Results indicated in shaded boxes are significantly different from the appropriate control. Hood, Light 55, and Cache Sl. at Lindsey Sl. were compared to the Low EC Control; Rough and Ready to the Mid EC Control; Suisun Slough at Rush Ranch to the High EC Control; Napa River at Vallejo Seawall to the High EC Control or Very High EC Control.

	Sampling Period:		3/17/09 - 3/19/09	3/31/09 - 4/2/09	4/14/09 - 4/16/09	4/28/09 - 4/30/09	5/12/09 - 5/14/09	5/26/09 - 5/28/09
	Age of Delta Smelt:		30 d	44 d	54 d	41 d	41 d	55 d
Endpoint	Treatment	Mean EC (µS/cm)	Survival					
96-h Survival	Low EC Control	160	NA	85.0	84.7^{N}/65.0^{A}	79.2^{N}/88.2^{A}	76.4	79.2
	Low EC/Low Turbidity Control	186	NA	66.8	46.7	92.5	68.8	87.5
	Low EC/Low Turbidity Control with Tannins	174	NA	31.8	-	-	-	-
	Hood	157	NA	51.0*	67.0*	79.5	62.9	89.7
	Light 55	262	NA	69.3	71.4	85.0	84.7	91.9
	Cache Lindsey	234	NA	53.6*	55.3	82.5	94.7	91.3
	Mid EC Control	644	NA	81.4	75.6	88.0	80.3	70.8
	Rough and Ready Island	593	NA	43.0	59.8	90.7	56.7	86.1
	High EC Control	3751	NA	86.1	82.5	100.0	86.4	92.5
	Low Turbidity Control	3750	NA	81.6	83.3	88.6	85.4	92.5
	Suisun	3672	NA	97.7	94.7	97.5	80.4	89.2
	Very High EC Control	15776	NA	-	-	-	72.1	70.8
	340	15078	NA	88.6	62.2**	97.7	68.9	67.5

* These samples showed significantly lower survival compared to an EC-specific control, but not compared to an EC- and turbidity-specific control.

** Significantly reduced survival was potentially caused by extremely high conductivity.

A Antibiotics added. Antibiotics were added to all treatments in tests initiated 4/30/09 and later.

N No antibiotics added.

Table 12, cont. Survival of larval *H. transpacificus* in water samples collected from the Sacramento - San Joaquin Delta. Results indicated in shaded boxes are significantly different from the appropriate control. Hood, Light 55, and Cache Sl. at Lindsey Sl. were compared to the Low EC Control; Rough and Ready to the Mid EC Control; Suisun Slough at Rush Ranch to the High EC Control; Napa River at Vallejo Seawall to the High EC Control or Very High EC Control.

7-d Survival	Low EC Control	160	8.3	70.0	58.9^{N} / 65.0^{A}	69.4^{N} / 85.9^{A}	71.4	76.4
	Low EC/Low Turbidity Control	186	2.8	43.0	27.4	85.2	59.7	75.0
	Low EC/Low Turbidity Control with Tannins	174	-	2.5	-	-	-	-
	Hood	157	8.7	19.5*	30.1*	55.3	52.3	71.1
	Light 55	262	23.6	40.7*	55.8	80.2	85.5	86.9
	Cache Lindsey	234	2.8	25.0*	46.9	67.5	80.1	81.3
	Mid EC Control	644	15.3	69.5	67.5	76.4	71.9	62.8
	Rough and Ready Island	593	2.8	9.3	42.2	88.2	28.1	72.8
	High EC Control	3751	18.6	64.5	70.0	100.0	80.8	82.5
	Low Turbidity Control	3750	18.1	61.6	61.9	86.1	55.2	71.4
	Suisun	3672	95.0	95.5	92.2	93.1	85.7	86.4
	Very High EC Control	15776	-	-	-	-	62.5	68.1
	340	15078	88.8	74.8	62.2	88.2	63.9	62.5

* These samples showed significantly lower survival compared to an EC-specific control, but not compared to an EC- and turbidity-specific control.

** Significantly reduced survival was potentially caused by extremely high conductivity.

A Antibiotics added. Antibiotics were added to all treatments in tests initiated 4/30/09 and later.

N No antibiotics added.

III.4 *In Situ* Monitoring on the Sacramento & San Joaquin Rivers

During March – May 2009, *in situ* monitoring was conducted at the DWR water quality monitoring stations located in Hood, CA (Sacramento River) and Rough & Ready Island in Stockton, CA (San Joaquin River). Six exposures using *H. transpacificus*, *P. promelas*, and *H. azteca* were conducted concurrently with ambient delta smelt toxicity testing in the laboratory. During this pilot project, no toxicity was detected in the Sacramento River at Hood or the San Joaquin River at Rough and Ready Island. *H. transpacificus* survival was poor, overall, but generally higher in ambient water than in the control, potentially due to slightly higher water temperatures in the control system. *H. azteca* survival was consistently high in ambient water as well as controls throughout the testing period. *P. promelas* survival was variable in both the control and ambient water. Poor *P. promelas* survival in controls was attributed to the addition of algal paste to optimize turbidity conditions for delta smelt larvae. Additional information including system design and exposure methods are provided below.

III.4.1 Methods

III.4.1.1 System Design

In situ devices were installed inside DWR water quality monitoring stations located directly above the Sacramento River in the town of Hood, CA and next to the San Joaquin River on Rough & Ready Island in Stockton, CA. Positioning the devices inside these small buildings had several advantages over placing the replicate cages inside the river itself, including improved temperature control, flow control, and ease of daily access. The device located at Rough & Ready Island was slightly different in layout than the device at Hood due to space restrictions, but overall function was the same. Ambient water was supplied from DWR's sampling station pump and delivered to the exposure chamber at 3.8 liters per minute (LPM). The apparatus consisted of three main parts: the ambient exposure chamber, the control exposure chamber, and the control sump. Plumbing that connected these three parts consisted primarily of common polyvinyl chloride (PVC) plumbing supplies. The function of each main part is described below.

The ambient exposure chamber consisted of a customized, white acrylic tank surrounded by an outer bath filled with flowing ambient water to maintain temperature. During the acclimation period for delta smelt, the chamber was filled with control water supplied from the control sump below, and at test initiation, control water was switched over to ambient water and the outer bath was drained. Held within the chamber were four replicate cages for each of the three test species (Figures 2, 3). The largest cages, used for larval delta smelt, *H. transpacificus*, were made from one gallon high density polyethylene (HDPE) buckets. These buckets and lids were black to provide optimal lighting conditions (less than 1 ft-candle through a hole in the lid) for *H. transpacificus*. Cages used for *P. promelas* and *H. azteca* were constructed from two manufactured parts; a low density polyethelene pipe cap (Niagra, Erie, PA) and nylon tea strainer (The Republic of Tea, Navato, CA). The exposure chamber lid that covered these cages was constructed from clear acrylic in order to allow ambient light into the chamber (16:8 light:dark cycle).

The control exposure chamber, exposure cages and lids were identical to those in the ambient system. Control water was supplied from the control sump immediately below and the control exposure chamber was also surrounded by an outer ambient water bath in order to maintain the temperature within 1 °C of the ambient water at all times. Flow was set at 3.8 LPM.

The control sump consisted of an 11 gallon HDPE bath containing a 210 gallons per hour (GPH) pond pump, which supplied recirculating control water to the control exposure chamber at all times, and to the ambient exposure chamber during acclimation only. The control water consisted of hatchery water diluted with deionized water or salted up with Instant Ocean to the same specific conductance as its corresponding ambient water. Approximately half the control water was replaced daily to reduce an accumulation of total ammonia in the control system and the control sump was aerated to ensure that dissolved oxygen levels remained at or near saturation.

III.4.1.2 Exposure Experiments

Diluted hatchery control water was used for acclimation of test organisms in the laboratory, and their transport to the DWR Station at Hood, as well as for the control treatment. Nanno 3600 Instant Algae (ReedMariculture, Inc., Cambell, CA) was added to increase the turbidity to 10 NTU to provide optimal conditions for *H. transpacificus.*

H. transpacificus (45-55 d old) were obtained from the UC Davis FCCL in Byron, CA. Fish were transported directly from the hatchery to each experimental site. Upon arrival, the fish were loaded into replicate buckets containing SC-adjusted hatchery water that matched their rearing conditions. The acclimation water also contained Nanno 3600 Instant Algae (ReedMariculture, Inc., Cambell, CA) to raise the turbidity to a minimum of 6 NTU. Over the course of the next 48 hours, the conductivity of the hatchery water was lowered slowly by adding deionized water or diluted hatchery water, until the conductivity matched that of the ambient water. *H. transpacificus* were fed *Artemia* nauplii three times daily during acclimation and once daily during the exposure.

Adult *H. azteca* were obtained from in house cultures and were acclimated in the lab for a minimum of 48 hours prior to the event. *P. promelas* were obtained from Aquatox, Inc. (Hot Springs, AR) and were acclimated a minimum of 24 hours prior to the event then deployed in the *in situ* exposure at 7 days old. A piece of dryed and leached leaf, measuring one cm squared, was placed into each *H. azteca* replicate cage prior to test initiation. All *in situ* species were fed once daily during the exposure period. *P. promelas* and *H. azteca* survival was recorded prior to test initiation and each day during the exposure. *H. transpacificus* survival was recorded at test initiation, on day 4, and at test termination due to the limited visibility in replicate buckets and the need to minimize disturbance.

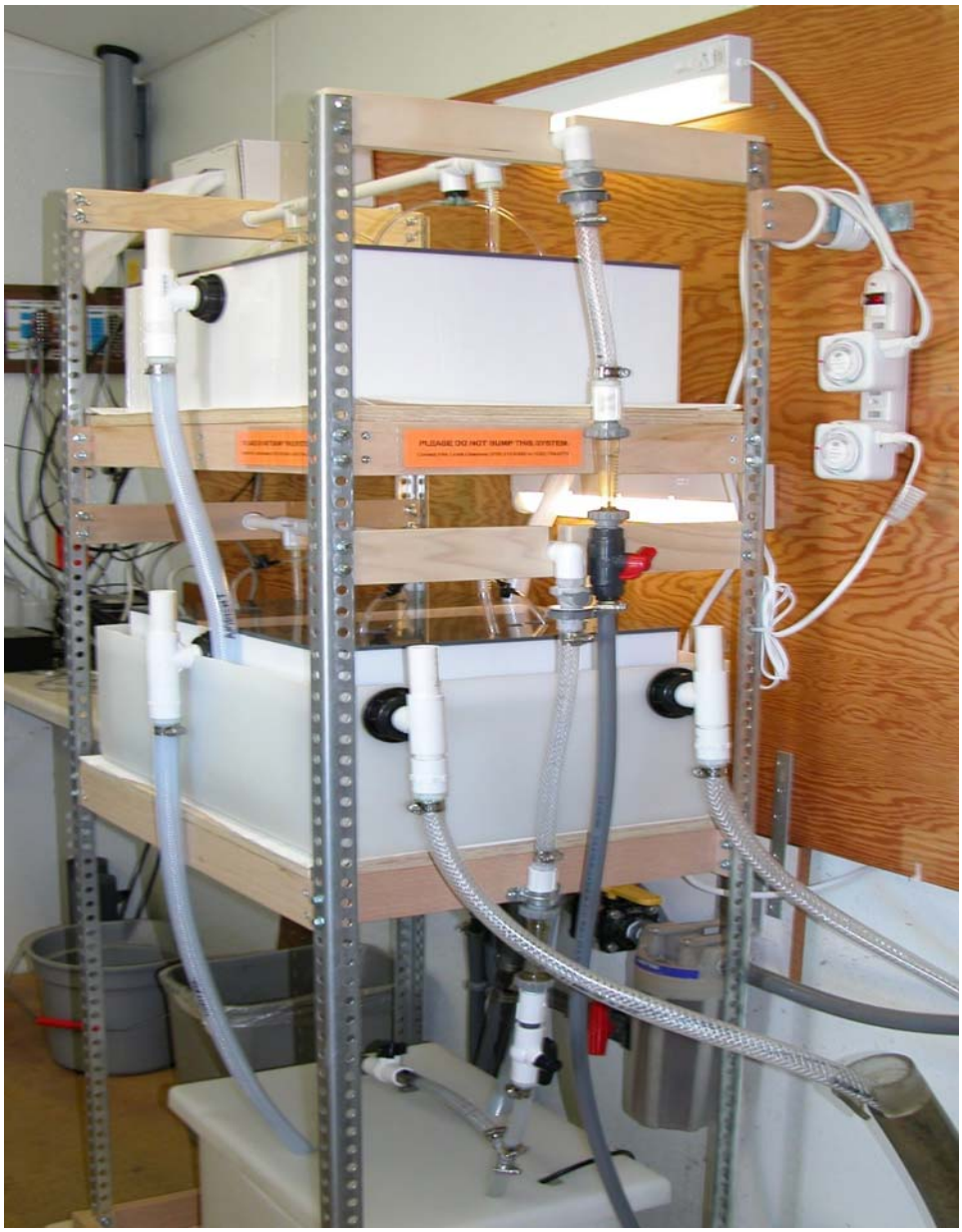

Figure 2. The in-situ exposure system at Hood.

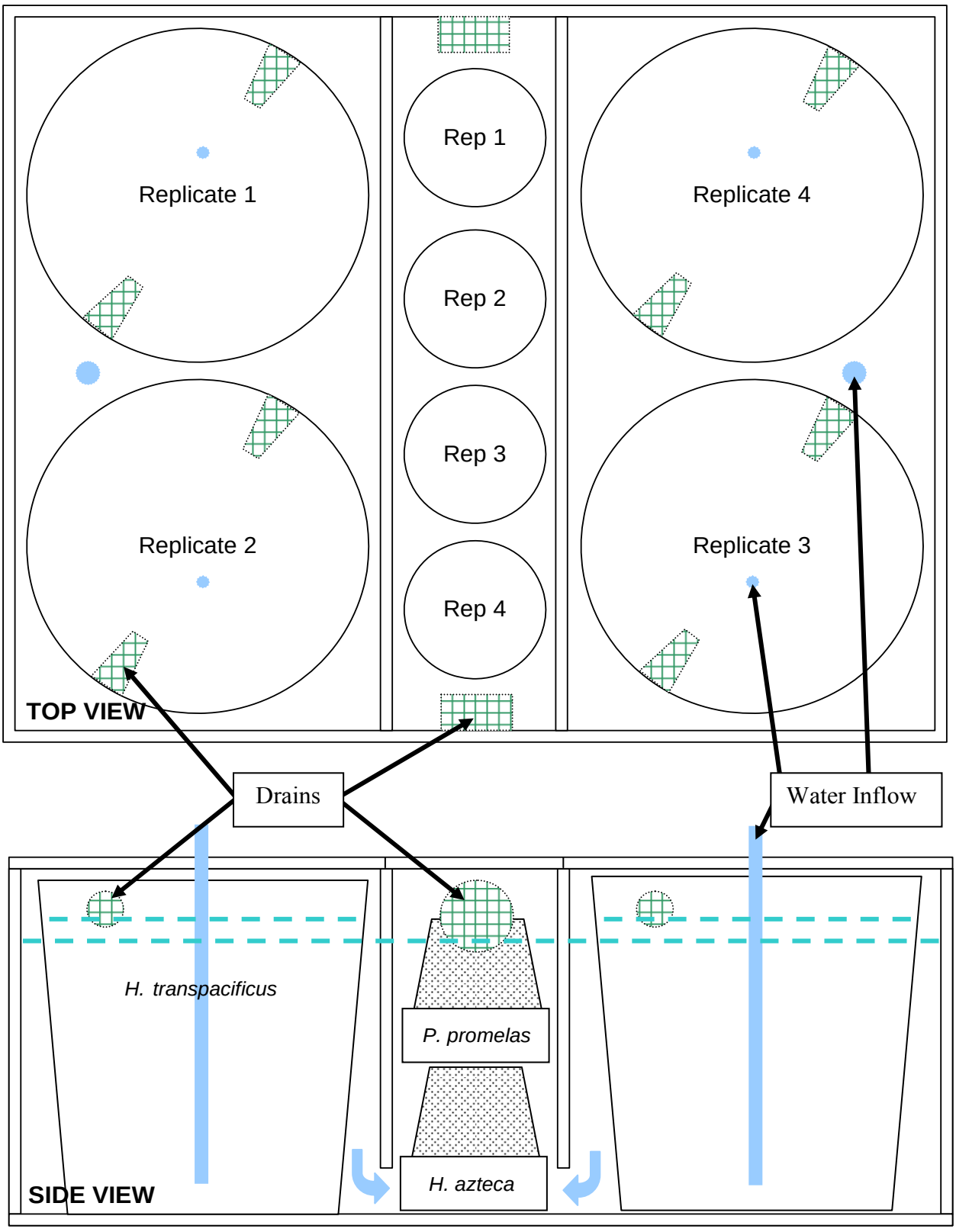

Figure 3. Top and side view of an exposure chamber for *in situ* devices.

Turbidity, temperature, total ammonia, pH, DO, SC, EC, hardness and alkalinity were measured in both the ambient and control exposure chambers daily. Once water was inside the exposure tanks, sediment did settle out to some degree causing an increase in sedimentation over the course of the experiment. Turbidity was also measured at the ambient water source to determine the turbidity going into the system. To the extent possible, SC, turbidity, and temperature were manipulated in the control to parallel the ambient exposure system. The SC and turbidity of the control water was adjusted daily immediately following a partial water exchange. Although we intended to adjust the turbidity of the control water to match the ambient water, we were unable to match the turbidity since the addition of too much alga confounds exposure results by increasing ammonia and producing more pathogens. Turbidity readings were consistently lower in the control water than the ambient water.

4.1.3 Statistical Analysis

Survival of each species was compared between control and ambient treatments using USEPA standard single-concentration statistical protocols.

4.2 Results

Tables 13 and 14 show the survival of *H. transpacificus*, *P. promelas* and *H. azteca* at the Rough and Ready DWR Station at Stockton and the Hood DWR Station on the Sacramento River. Detailed data tables are provided in Appendix D. No significant reductions in survival were detected at either site during any experimental event. *H. transpacificus* survival was generally higher in ambient waters than in the controls, *H. azteca* survival was consistently high throughout the *in situ* season, and *P. promelas* survival was variable in both the control and ambient waters.

Table 13. 96-hour and 7-day survival of animals examined in flow-through *in situ* tests initiated at the Rough and Ready DWR Station, Stockton, CA.

Date	Treatment	*H. transpacificus*				*P. promelas*				*H. azteca*			
		96-h Survival (%)		7-d Survival (%)		96-h Survival (%)		7-d Survival (%)		96-h Survival (%)		7-d Survival (%)	
		mean	se	mean	se	mean	se	mean	se	mean	se	mean	se
3/19/2009	Control	-	-	22	10.4	94	6.3	71	16.1	95	5.0	95	5.0
	Ambient	-	-	35	9.3	80	0.0	65	9.6	100	0.0	100	0.0
4/2/2009	Control	61	8.9	41	7.9	60	8.2	45	5.0	95	5.0	95	5.0
	Ambient	75	6.8	61	9.4	90	10.0	90	10.0	100	0.0	100	0.0
4/16/2009	Control	63	9.7	59	12.4	65	12.6	65	12.6	100	0.0	100	0.0
	Ambient	71	9.8	66	12.5	45	12.6	40	8.2	100	0.0	100	0.0
4/30/2009	Control	79	12.5	68	15.8	75	9.6	70	12.9	95	5.0	90	10.0
	Ambient	61	16.5	47	14.1	70	12.9	70	12.9	100	0.0	85	9.6
5/14/2009	Control	15	9.6	0	0.0	95	5.0	95	5.0	100	0.0	95	5.0
	Ambient	15	8.6	15	8.6	100	0.0	100	0.0	100	0.0	100	0.0
5/28/2009	Control	-	-	-	-	100	0.0	100	0.0	100	0.0	100	0.0
	Ambient	-	-	-	-	100	0.0	100	0.0	100	0.0	95	5.0

Table 14. 96-hour and 7-day survival of animals examined in flow-through *in situ* tests initiated at the DWR Station on the Sacramento River at Hood.

Date	Treatment	*H. transpacificus*				*P. promelas*				*H. azteca*			
		96-hr Survival (%)		7-day Survival (%)		96-hr Survival (%)		7-day Survival (%)		96-hr Survival (%)		7-day Survival (%)	
		mean	se	mean	se	mean	se	mean	se	mean	se	mean	se
3/19/2009	Control	-	-	21	5.5	100	0.0	85	15.0	100	0.0	95	5.0
	Ambient	-	-	46	8.4	85	9.6	75	15.0	95	5.0	95	5.0
4/2/2009	Control	75	4.8	62	8.8	85	9.6	30	5.8	95	5.0	80	8.2
	Ambient	84	10.3	77	7.0	90	5.8	85	5.0	85	9.6	80	8.2
4/16/2009	Control	59	5.0	29	5.1	95	5.0	95	5.0	95	5.0	90	5.8
	Ambient	74	10.5	64	13.8	90	5.8	85	9.6	95	5.0	85	9.6
4/30/2009	Control	47	10.9	43	13.3	95	5.0	95	5.0	100	0.0	100	0.0
	Ambient	43	6.5	40	6.9	100	0.0	95	5.0	100	0.0	100	0.0
5/14/2009	Control	56	18.8	44	15.7	95	5.0	95	5.0	100	0.0	100	0.0
	Ambient	69	12.0	50	10.2	100	0.0	100	0.0	100	0.0	100	0.0
5/28/2009	Control	13	8.0	4	4.2	85	5.0	85	5.0	95	5.0	95	5.0
	Ambient	34	7.9	27	8.4	95	5.0	85	15.0	100	0.0	90	5.8

III.4.3 Discussion

One of the greatest advantages to the *in situ* exposure is that the organisms experience the fluctuations of toxicant concentrations for the same length of time that stationary organisms in the river would experience them. In contrast, organisms that are exposed in a laboratory setting to a one-time grab sample experience the same water sample for a defined test period. A one-time grab sample can be collected when the concentrations of a chemical are at its peak, fall well below the peak concentration or miss a chemical pulse entirely. Laboratory static renewal tests utilizing one-time sub surface grab samples can therefore overestimate or underestimate toxicity depending on when a sample is collected relative to a toxic pulse moving through the system. The in situ devices renew water continuously with approximately 95% of the water renewed every half hour. The constant flow to the system is representative of the river conditions throughout the exposure period.

No toxicity was detected in the Sacramento River at Hood or the San Joaquin River at Rough and Ready Island suggesting that any toxicant(s) that may have traveled through the system were not at high enough concentrations for enough time to cause reduced survival to the test species. *H. transpacificus* survival was generally higher in ambient water than in the control, which decreased our ability to detect a toxic event with the species. A number of variables, including natural food supply, temperature, and turbidity may have contributed to higher delta smelt survival in ambient water compared to the controls. *H. azteca* survival was consistently high in ambient water and control water while *P. promelas* survival was variable in both, possibly due to the promotion of bacterial growth following the addition of *Nannochloropsis*.

Despite our efforts to slowly acclimate the *H. transpacificus* to the conductivity and temperature conditions of river water at *in situ* sites, survival of delta smelt remained low. Our recommendations are to use a test species that is more tolerant of transport, salinity and temperature stresses. *P. promelas* and *O. mykiss* appear to be far more tolerant of such stressors. *O. mykis*s might be a suitable species to use during the cold months and a warmwater species might be more suitable during the warmer months.

III.5 Toxicity Monitoring with Copepods

III.5.1 Methods

III.5.1.1 Collection and Culturing

E. affinis and *P. forbesi* were cultured at the UCD-ATL after being collected from the field or obtained from the Romberg Tiburon Center, Tiburon, CA. When copepods were collected in the field, 5-minute tows were performed using a 147 µm plankton net, either by boat, from a dock, or from the bank. Following each tow, the collection tube was emptied into a 1-gal plastic bucket filled with site water and then visually inspected to determine the density of zooplankton. This was done after each tow in order to avoid stress and mortality induced by overcrowding. The 1-gal buckets were then placed in a cooler and transported to UCD-ATL where they were immediately transferred to an environmental chamber adjusted to the ambient water temperature of the collection site. Dates, locations, and conditions of field collections are summarized in Table 15.

Table 15. Copepod collections in the Delta during October 2009 – February 2010.

Collection Date	Location	Temp. (°C)	SC (µS/cm)	Catch
10/16/2009	Antioch	17.3	1466	Many cyclopoids and non-target calanoids. *P. forbesi* and *E. affinis present*.
12/1/2009	Antioch	10.5	2309	Many cyclopoids and non-target calanoids. *E. affinis present*.
12/21/2009	Rio Vista	9.5	242	Cyclopoids abundant. *E. affinis* present. Very few *P. forbesi*.
1/6/2010	Rio Vista	9.0	251	Cyclopoids abundant. *E. affinis* and *P. forbesi* present.
1/15/2010	Antioch	9.4	1923	*E. affinis* and cyclopoids abundant in collection. Very few *P. forbesi*.
2/2/2010	West Sacramento	10.5	995	*P. forbesi* abundant. Cyclopoids and non-target calanoids present. *E. affinis present.*
2/18/2010	West Sacramento	14.0	957	Cyclopoids and non-target calanoids abundant. Many *P. forbesi* and some *E. affinis.*

Following collection and transport to the laboratory, *P. forbesi* and *E. affinis* were isolated and identified using a dissecting scope and depression slides. The desired copepods were gently pipetted into 250 ml glass beakers containing 1 µm filtered site water collected at the time of sampling. These beakers were returned to the environmental chamber and moved into larger culturing vessels at the end of the day.

E. affinis copepodites obtained from the Romberg Tiburon Center were placed in a 1-gal plastic jar containing culture water and transported to UCD ATL in a cooler. Upon receipt, the organisms were placed in an environmental chamber and acclimated to test conditions. Cultures were maintained in either 5-gal acrylic cones or 4-L glass beakers, depending on the number of organisms isolated. Density was maintained at 25-50 adult copepods/L and organisms were fed diluted Shellfish Diet daily. Culture vessels were cleaned and approximately 50% of the culture water was replaced weekly.

In preparation for toxicity testing, juvenile or adult copepods were selected by inspecting sub-samples of cultures under a dissecting scope and pipetting animals individually.

III.5.1.2 Disease and Treatment

In several batches of field collected copepods, *E. affinis* was found to be infested to varying degrees with a parasitic ciliate called *Vorticella*. These ciliates attach themselves by a slender stalk to the bodies of the copepods, and use them as a vehicle to obtain food including bacteria and small protozoans. *Vorticella* do not affect copepod health unless food supply is limited and parasites are competing with their hosts for food.

Infestations were controlled by removing infected copepods from the cultures to prevent the ciliates from spreading. In addition, experimental treatments were added to the *E. affinis* copper reference toxicant test initiated on January 31, 2010 to determine if the infestation could be treated with 1 ppb copper, and if heavily infested animals could survive for the duration of the 96-h test. The copper treatment had no effect, but the data showed that copepod survival was not compromised by heavy *Vorticella* infestation .

No diseases were noted or treated in *P. forbesi*.

III.5.1.3 Toxicity Testing

Copepods were acclimated to test conditions for 48 h prior to testing. Before initiating bioassays, the water samples were mixed rigorously in the original sampling containers, filtered through a 60-µm screen, brought to test temperature of 15 ºC and aerated at a rate of 100 bubbles/minute until the DO concentration was approximately 9.5 mg/L. Sierra Springs™ water amended to US EPA moderately hard standards (SSEPAMH) and adjusted to an SC of 500µS/cm was used as the primary laboratory control water. Each series of tests included a standard laboratory control, and if necessary, a "low EC control". "Low EC" control water was reconstituted to US EPA moderate hardness and the EC was adjusted to match the lowest EC of the water samples by diluting with glass distilled water. During Event 2, *E. affinis* were taken from cultures held at a salinity of 8 ppt. These animals were acclimated to 2 ppt, and both controls and ambient waters were amended to 2 ppt for testing.

The 96-hour tests consisted of ten 20 ml replicate glass scintillation vials, each containing 15 ml of sample and 1 organism. Tests were initiated with juvenile copepods whenever possible, and if juveniles were not available, adult copepods were used. Each replicate vial was fed 15 µl of diluted Shellfish Diet (Reed Mariculture, Campbell, CA) at test initiation and during the daily renewal of 75% of test waters. Tests were conducted at a temperature of 15 ± 2 ºC with a 16h:8h Light:Dark photoperiod. Mortality was recorded daily. Details of *E. affinis* and *P. forbesi* test conditions are given in Tables 16 and 17.

Table 16. Test conditions and protocols for 96-h *E. affinis* test.

Test method: *Eurytemora affinis* 96-h Water Column Initial Screening Test **Method number:** Modified from method number 1002.0 (UCD ATL SOP 1-7, non-certification) **Method source:** Adapted from the Chronic *Ceriodaphnia dubia* testing protocol outlined in Methods for Estimating the Chronic Toxicity of Effluents and Receiving Waters to Freshwater Organisms, 4th ed., 2002, EPA/821/R-02/013. **Endpoints:** 96-hour Survival **Test chamber type and volume:** 20 mL glass scintillation vial **Volume of sample per chamber:** 15 mL **Organisms per chamber:** 1 **Replicates per treatment:** 10 **Acclimation of test organisms:** 48 hours prior to start of the test **Renewal:** 75% daily renewal **Feeding frequency, amount and type:** 15 µl of Dilute Shellfish Diet per replicate daily **pH control measures:** None; pH between 6-9 **Aeration:** None; treatment waters aerated if supersaturated prior to renewal

Table 17. Test conditions and protocols for 96-h *P. forbesi* test.

Test method: *Pseudodiaptomus forbesi* 96-h Water Column Initial Screening Test **Method number:** Modified from method number 1002.0 (UCD ATL SOP 1-8, non-certification) **Method source:** Adapted from the Chronic *Ceriodaphnia dubia* testing protocol outlined in Methods for Estimating the Chronic Toxicity of Effluents and Receiving Waters to Freshwater Organisms, 4th ed., 2002, EPA/821/R-02/013. **Endpoints:** 96-hour Survival **Test chamber type and volume:** 20 mL glass scintillation vial **Volume of sample per chamber:** 15 mL **Organisms per chamber:** 1 **Replicates per treatment:** 10 **Acclimation of test organisms:** 48 hours prior to start of the test. **Renewal:** 75% daily renewal **Feeding frequency, amount and type:** 15 µl of Dilute Shellfish Diet per replicate daily **pH control measures:** None; pH between 6-9 **Aeration:** None; treatment waters aerated if supersaturated prior to renewal

III.5.1.5 Statistical Analysis

Data was analyzed using USEPA standard single-concentration statistical protocols (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. Each comparison of a sample to a control was treated as a separate statistical test, in accordance with USEPA (2002).

III.5.2 Results

In the control series, survival was best in the highest conductivity treatment of 1900 µS/cm (90% survival after 7 d) and decreased with decreasing conductivity. Survival was generally low in ambient samples with the exception of Cache-Ulatis (100% survival after 7 d). This sample had the highest turbidity (45.9 NTU) and specific conductance (329 µS/cm) of all four sites which may have contributed to better animal performance, despite the low survival encountered in the corresponding conductivity control (Table 18). Detailed data tables are provided in Appendix E. Survival in all ambient samples was higher than survival in the corresponding control water, however it is apparent that conductivity was the most important factor determining copepod survival in all samples tested.

Table 18. Results of a *E. affinis* 7-d test initiated 5/1/09 evaluating the toxicity of ambient samples collected on 4/28/09 and 4/30/09.

Treatment	Measured Specific Conductivity (uS/cm)	Survival (%)[1]	
		Mean	SE
L16 Media @ 1 ppt	1930	90	10.0
L16 Media @ 1000 µS/cm	1003	50	16.7
L16 Media @ 500 µS/cm	517	30	15.3
L16 Media @ 250 µS/cm	282	20	13.3
L16 Media @ 100 µS/cm	129	0	0.0
Sacramento R. Deep Water Channel, Light 55	271	50	22.4
Sacramento River at tip of Grand Island (711)	136	20	13.3
Upper Cache Slough at mouth of Ulatis Creek	329	100	0.0
Sacramento River at Hood DWR Station	142	20	13.3

1. Highlighted cells indicate statistically significant reductions in survival compared to the L16 media @ 1 ppt. Ambient samples showed no significant decreases in survival compared to the most appropriate conductivity control waters. Data were analyzed using USEPA standard statistical protocols.

III.6. Quality Assurance / Quality Control

Quality Assurance/Quality Control (QA/QC) measures are included in this project to assess the reliability of the data collected. These QA/QC procedures include positive control tests (i.e., reference toxicant tests), field duplicates, bottle blanks and trip blanks. Duplicate, bottle and trip blank samples were selected from *H. azteca*-specific sampling sites because QA/QC comparisons were not included in the developmental *H. transpacificus* bioassays. The components of these QA/QC measures are outlined below.

Reference toxicant tests: Positive control tests (reference toxicant tests) are conducted to ascertain whether organism responses fall within the acceptable range as dictated by US EPA. The LC_{50}/EC_{25} for each reference toxicant endpoint was plotted to determine whether it fell within the 95% confidence interval of the running mean. If the LC_{50}/EC_{25} falls out of the 95% confidence interval, or plus or minus two standard deviations around a running mean, test organism sensitivity is considered atypical and results of toxicity tests conducted during the month of reference toxicant outliers may be considered suspect. Reference toxicant tests with *H. azteca, C. dubia, P. promelas* and *O. mykiss* were performed using sodium chloride as the toxicant. Reference toxicant tests with *H. transpacificus* and *E. affinis* were conducted with copper (II) chloride as the toxicant.

H. azteca: Reference toxicant tests were conducted monthly. From January 1, 2008 to December 31, 2009, *H. azteca* performed normally within each reference toxicant test.

C. dubia: Chronic reference toxicant tests were performed monthly. An acute reference toxicant test was performed in March, 2010, to coincide with acute LC_{50} toxicity testing. Chronic and acute *C. dubia* performance was within acceptable limits during the project period.

P. promelas: Reference toxicant tests were performed monthly to coincide with LC_{50} toxicity testing. *P. promelas* performance was within acceptable limits during the project period.

O. mykiss: One reference toxicant test was performed concurrently with a toxicity test initiated on November 11, 2008, which examined toxicity observed in the field at Rough and Ready Island, DWR Station, Stockton. Organisms in the reference toxicant test performed normally.

H. transpacificus: Four successful reference toxicant tests were conducted during the study period; organism performance was within acceptable limits.

E. affinis: Reference toxicant tests were conducted monthly between January and March, 2010. Two reference toxicant tests did not meet the test acceptability criterion for survival in March, due to poor organism health. However, although survival in the controls in these tests was less than the required 75%, effect concentration data was obtained.

Field duplicates: Field duplicate samples were collected to assess precision. Field duplicate samples are in agreement when the primary sample and its duplicate are

both either statistically similar or statistically different from the control. Forty-eight field duplicates were collected during 2008-2010 of the study. Of those forty-eight samples, 100% shared equivalent survival results and 98% shared equivalent weight results. The frequency of field duplicates sharing equivalent results is outlined in Table 19.

Bottle blanks: Bottle blank samples were included to evaluate potential incidental contamination due to the sample container. Bottle blanks are analyte-free water samples that are transferred to a clean sample container that is prepared in the laboratory. Bottle blanks were comprised of DIEPAMHR. A bottle blank sample is in agreement when it is statistically similar to the control. Thirty bottle blanks were analyzed during 2008-2010 of the study. Of those thirty samples, 100% shared equivalent survival results and 92% shared equivalent weight results. The frequency of bottle blank samples sharing equivalent results is outlined in Table 19.

Trip blanks: Trip blank samples were included in this project 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. Trip blank samples were comprised of DIEPAMHR. A trip blank sample is in agreement when it is statistically similar to the control. Twenty-four trip blank samples were analyzed during 2008-2010 of the study. Of those twenty-four samples, 100% shared equivalent survival and weight results. The frequency of trip blank samples sharing equivalent results is outlined in Table 19.

Table 19. Frequency of QA/QC samples sharing equivalent results

QA/QC Samples	Sample Size	% Agreement	
		Survival	Weight
Field Duplicates	48	100	98
Bottle Blanks	30	100	92
Trip Blanks	24	100	100

Precision: Precision is the degree to which the primary sample agrees with its duplicate. Precision can be measured by calculating the Relative Percent Difference (RPD) between sample measurements. The RPD between a sample and its duplicate can be calculated by using the following equation:

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

RPDs were calculated using the aforementioned equation on water chemistry measurements such as DO, pH, EC, hardness and alkalinity and ammonia. Both the individual and average RPDs between duplicates are outlined in detail in Appendix K. Caution should be applied when interpreting water quality precision data. Often times an

unusually high RPD is due very minute amounts of a particular analyte being measured rather than a lack of precision.

Deviations: Six deviations occurred during the 2008-2010 project study period. Deviations consisted of warm sample temperatures at receipt, exceeded holding times, and a protocol deviation. Frequency of deviations is outlined in Table 20.

Sample temperatures: USEPA requires sample temperatures to be maintained between 0-6 ºC during transport, receipt and storage. Elevated sample receiving temperatures occurred during the summer months of the study period and were due to ice melting in the transport coolers. Samples in which temperatures exceeded the EPA criterion were within half a degree of the target temperature of 6 ºC and were placed in the dark in cold storage immediately after receipt. These samples performed normally in toxicity tests and the data associated with these samples are considered reliable.

Holding times: Two *H. azteca* tests conducted during the study period were initiated past the 72-hour holding time. These tests were a retest of the original samples which were initiated in a toxicity test which did meet proper holding time requirements. One toxicity test initiated on August 28, 2008 did not meet test acceptability criteria and had to be repeated. The second toxicity test, initiated on May 14, 2009, exhibited contamination related to the PBO-manipulated samples and had to be repeated. The retests met all test acceptability criteria and the data are considered reliable.

Protocol deviation: An *H. azteca* toxicity test initiated on August 29, 2008 was initiated with a reduced number of animals per replicate. This deviation occurred because there was a shortage of healthy animals available for test initiation. In order to meet holding time requirements, the test was initiated using only the healthiest animals in the culture, which resulted in having a reduced number of organisms in each replicate. This test met all test acceptability criteria and the data are considered reliable.

Table 20. Frequency of deviations

Deviation Type	Date of Deviation	Explanation
Exceeded sample temperature	CL 5/8/2008 602, 609 4/1/2009 887 (Site 915 FD) 8/6/2009	Ice melting in transport coolers
Exceeded holding time	*H. azteca* retest initiated 9/10/2008 *H. azteca* retest initiated 5/16/2009	Test did not meet test acceptability criteria PBO contamination
Protocol deviation	*H. azteca* test initiated 8/29/2008	Shortage of healthy animals available; test initiated with reduced number of animals per replicate

Completeness: UCD ATL strives for a 90% completeness of work performed. The following is a summary of work completed for the duration of the study period of

2008-2010. Table 21 denotes the number of successful tests completed out of number of tests conducted. Please note there are no test acceptability criteria for *in-situ* testing.

Table 21. Completeness during the project period 2008-2010.

Species	Test	Number of Tests	% Completeness
H. azteca	Ambient	99 / 101 (2 tests repeated)	100
	LC_{50}	7 / 7	100
	In-situ	6 / 6	100
C. dubia	LC_{50}	6 / 6	100
P. promelas	LC_{50}	7 / 7	100
	In-situ	6 / 6	100
H. transpacificus	Ambient	7 / 10	70
	LC_{50}	7 / 7	100
	In-situ	6 / 6	100

III.7 References

California State Water Resources Control Board. 2002. Toxicity Testing SOPs: Hyalella azteca 10-Day Water Toxicity Test. Quality Assurance Management Plan for the State of California's Surface Water Ambient Monitoring Program. Division of Water Quality. Sacramento, CA.

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IV. Comparative Sensitivity of Resident Species to Chemical Stressors

The lack of information on the toxic effects of contaminants on resident Delta species, among them delta smelt and important prey species, prevents an estimation of the risk of chemical contamination to pelagic organisms of concern. This study generated comparative sensitivity data (in the form of 96-h LC50, EC50, no observed effect level (NOEC), and lowest observed effect level (LOEC) for Delta species and standard toxicity test species. Testing included *Pseudodiaptomus forbesi*, *Eurytemora affinis, Ceriodaphnia dubia*, *Hyalella azteca*, delta smelt, and fathead minnow.

Chemicals were selected based on their relevance for the Delta: copper, ammonia, the organophosphate insecticides chlorpyrifos and diazinon, and the pyrethroid insecticides, cyfluthrin, bifenthrin, permethrin. Copper is used as a pesticide in various forms, is a common chemical in stormwater runoff, and is ubiquitous in the aquatic environment. Ammonia is released from wastewater treatment plants. Chlorpyrifos is one of the most heavily used agricultural insecticides, and has recently been shown to be present at toxic concentrations in Ulatis Creek (Werner and Kuivila, 2004, unpublished data) and agricultural drains (California Regional Water Quality Control Board Agricultural Waiver Program, 2007). Diazinon, bifenthrin, cyfluthrin, and permethrin were detected in 2007 in water column samples from various sites in the Delta (Werneret al., 2008).

Samples from each pesticide concentration as well as a control were submitted to CDFG-WPCL to verify nominal chemical concentrations. In tests evaluating toxicity in both control and hatchery waters, only samples of hatchery water were submitted for chemical analysis. Total ammonia measurements for the ammonia chloride tests were measured at the UCD ATL. Sensitivity testing methods for each species are described below.

IV.1 Methods

Rangefinder tests were conducted to assist the selection of exposure concentrations for the "definitive" LC50/EC50 tests. Rangefinder test methods followed protocols described by US EPA (2002 a, b), but were limited to two replicates per treatment and a duration of 48 h. Water quality parameters (electrical conductivity (EC, dissolved oxygen concentration (DO), pH, ammonia concentration, hardness and alkalinity) were measured at test initiation; pH, temperature and DO were measured daily before and after water renewal.

IV.1.1 *H. transpacificus* 96-h Survival

For definitive LC50 tests, *H. transpacificus*, ranging from 39 to 51 days post hatch (DPH), were obtained from the UC Davis FCCL in Byron, CA. The organisms were acclimated a minimum of 24 hours with hatchery water adjusted to a specific conductance (SC) of 900 µS/cm using Instant Ocean and a pH of 7.9 using HCl. *H. transpacificus* were fed *Artemia* nauplii three times daily during acclimation and during the exposures. After the acclimation period, ten organisms were randomly transferred into each of four replicate buckets using a 50 ml beaker.

Tests were performed in hatchery water (Appendix A, Table A-7) filtered through a 1 micron filter at 900 µS/cm and pH 7.9. Chemicals were dissolved in methanol (insecticides only) or water, and spiked into treatment solutions the morning the test was initiated and again on the renewal day. Tests were conducted in a water bath maintained at 16°C surrounded by dark shower curtains to minimize light-induced stress. One gallon black buckets with lids were used as replicate vessels, each containing 3.5 liters of sample. During testing, lids were allowed to rest on top of the buckets, but were not snapped shut to allow less than one ft-candle of ambient light in. Where methanol was used as a solvent, solvent control treatments containing 0.05% methanol (equal to the highest concentration added to insecticide treatments) were added. These methanol treatments were aerated after recognition of dissolved oxygen problems associated with the addition of the methanol, likely due to bacterial growth and associated respiration.

Mortality was recorded daily using a small flashlight. On day 2, 80% of test solutions were renewed during which dead fish, excess *Artemia* and other detritus were removed. At the end of the 96-h exposure, surviving organisms were euthanized with MS-222 and preserved with liquid nitrogen for subsequent molecular studies.

Water quality parameters (specific conductance (SC), temperature, dissolved oxygen concentration (DO), pH, total ammonium concentration, hardness and alkalinity) were measured at test initiation; in addition, pH, ammonium, temperature and DO were measured on day 2 before and after water renewal, and at test termination. Samples of each exposure concentration and a control were preserved and submitted to the CDFG analytical laboratory, Rancho Cordova, to verify nominal chemical concentrations. Additional analytical samples were taken following the first 48 hour exposure period to account for pesticide adsorption to container walls and food.

IV.1.2 *P. promelas* 7-d Survival and Growth

Larval fathead minnows were obtained from Aquatox, Inc. (Hot Springs, AR). Organisms used in sensitivity tests were <48 hours old and were acclimated to laboratory conditions 24 hours prior to test initiation.7-d LC50 test methods followed those outlined in the Fathead Minnow Larval Survival and Growth Test (USEPA, 2002). These tests were performed in deionized water amended to US EPA moderately hard standards (DIEPAMH) as well as hatchery water filtered through a 1 micron filter. Water was adjusted to an SC of 900 µS/cm using Instant Ocean and a pH of 7.9 using HCl.

Stock solutions were prepared by dissolving pesticides in methanol and ammonia and copper in glass distilled water. Chemicals were spiked into test solutions on Days 0, 2, 4, and 6. Where methanol was used as a solvent, solvent control treatments containing 0.05% methanol (equal to the highest concentration added to insecticide treatments) were added. These methanol treatments were aerated after recognition of dissolved oxygen problems associated with the addition of methanol, likely due to bacterial growth and associated respiration.

Mortality was recorded daily, and at test termination, a portion of organisms were preserved using liquid nitrogen for subsequent molecular studies while the rest were dried to a constant weight for the biomass endpoint. If ten surviving fish were present in a replicate at test termination, five were preserved with liquid nitrogen and five were dried;

if nine surviving fish were present in a replicate, 4 were preserved with liquid nitrogen and five were dried; if eight surviving fish were present in a replicate, four were preserved with liquid nitrogen and four were dried. If there were seven or less surviving fish in a replicate, all were dried to calculate biomass and average weight per individual.

Water quality parameters (specific conductance (SC), temperature, dissolved oxygen concentration (DO), pH, total ammonium concentration, hardness and alkalinity) were measured at test initiation; in addition, pH, ammonium, temperature and DO were measured on days 2, 4 and 6, before and after water renewal, and at test termination. Samples of each exposure concentration and a control were preserved and submitted to the CDFG analytical laboratory, Rancho Cordova, to verify nominal chemical concentrations.

IV.1.3 *H. azteca* 10-day Survival and Growth

H. azteca purchased from Aquatic Research Organisms were received at the UCD ATL 48 hours prior to test initiation and acclimated to laboratory conditions. The 10-day sensitivity tests were conducted in both DIEPAMHR (Appendix A, Table A-7) and "hatchery water" collected from the UCD FCCL. Waters were adjusted to a SC of 900 µS/cm using Instant Ocean and a pH of 7.9 using HCl. Prior to initiating bioassays, the water samples were brought to the test temperature of 23º C and aerated at a rate of 100 bubbles/min until the dissolved oxygen concentration was approximately 8.5 mg/L.

Sensitivity tests consisted of four 250 ml replicate glass beakers, each containing 100 ml of sample, a one-square-inch piece of nitex screen and 10 organisms. Tests were initiated with 7-14 day-old *H. azteca*. Animals in each replicate were fed 1000 µl of YCT on test initiation and on days 2, 4, 6 and 8, following the renewal of 75% of the test waters. Each series of sensitivity tests included a standard laboratory control, hatchery water control and any applicable method blanks.

Tests were conducted at a temperature of 23 ± 2º C with a 16h:8h, light:dark photoperiod. Mortality was recorded daily and waters were renewed on days 2, 4, 6 and 8. On day 10, the surviving *H. azteca* were dried and weighed to determine dry tissue weight per individual and relative growth. Effect data such as NOEC, LOEC, LC_{10}, LC_{50} and EC_{25} were calculated on both the 96-h and 10-d endpoints.

IV.1.4 *C. dubia* Tests

96-h Survival: Toxicity testing for acute *C. dubia* 96-h LC50 tests followed the static-renewal procedures outlined in "Methods for Measuring the Acute Toxicity of Effluents and Receiving Waters to Freshwater and Marine Organisms" (USEPA, 2002). The 96-h acute *C. dubia* test consists of four replicate 20 ml glass scintillation vials containing 18 ml of sample and five organisms. Tests are initiated with less than 24-h neonates, born within a 20-h window. Organisms are fed a mixture of YCT (a mixture of yeast, organic alfalfa and trout chow) and *S. capricornutum* two hours prior to daily water renewal in order to minimize adsorption of chemicals to food particles. *C. dubia* are transferred into a new vial of fresh solution daily. Sierra Springs (TM) drinking water amended to US EPA moderately hard standards (SDEPAMH; Appendix A, Table A-7))

is used as the control water for the *C. dubia* test. Treatment solutions were made daily and added in increasing concentrations to SDEPAMH, pH-adjusted to 7.9, and salinity-adjusted to 900 µS/cm with Instant Ocean. Tests were conducted at 25 ± 2ºC with a 16-h light: 8-h dark photoperiod. Mortality was measured daily and at test termination.

Seven-Day Survival and Reproduction: The *C. dubia* chronic test consists of ten replicate 20 ml glass vials each containing 15 ml of test solution and one organism. *C. dubia* are cultured at UCD ATL. Tests were initiated with less than 24-h old *C. dubia*, born within an 8-h period. *C. dubia* were fed a mixture of *S. capricornutum* and YCT (a mixture of yeast, organic alfalfa and trout chow) and transferred into a new vial containing fresh test solution daily. Sierra Springs™ water amended to EPA moderately hard specifications (SSEPAMH) was used as method control, in addition to the synthetic water. Tests were conducted at 25 ± 2 ºC with a 16-h light: 8-h dark photoperiod. Mortality and reproduction (number of neonates) were recorded daily and at test termination (after the third brood; day 6-8).

IV.1.5 *E. affinis* Tests

Please refer to Chapters III.5 and IV.3 (Teh et al., 2009) for a detailed description of *E. affinis* testing methods.

In addition to tests conducted in the laboratory of Dr. S. Teh, UC Davis, the UCD ATL conducted a series of tests to confirm earlier results obtained by Dr. Teh. Copper was used as a reference toxicant for a CV Regional Water Quality Control Board study measuring the toxicity of Delta island agricultural drains. The results of these tests are provided below. Rangefinder tests were conducted for a number of insecticides, but for lack of time, the respective LC50 tests were not completed. Nevertheless, results of rangefinder studies are also provided for comparison.

LC50 tests: The *E. affinis* 96-h LC50 tests consisted of ten 20 ml replicate glass scintillation vials, each containing 15 ml of test solution and 1 organism. Rangefinder tests used four replicate glass vials containing 15 ml of test solution and 1 organism. Tests were initiated with juvenile copepods whenever possible, and if juveniles were not available, adult copepods were used. Each replicate vial was fed 15 µl of diluted Shellfish Diet (Reed Mariculture, Campbell, CA) at test initiation and during the daily renewal of 75% of test waters. Tests were conducted at a temperature of 20 ± 2 ºC, a conductivity of 900 µS/cm, and pH 7.9 with a 16h:8h Light:Dark photoperiod. Mortality was recorded daily. Details of *E. affinis* and *P. forbesi* test conditions are provided in Chapter III.5.

Reference toxicant tests with copper: The sensitivity of *E. affinis* to copper was examined in reference toxicant tests performed multiple times. These tests were conducted using a synthetic laboratory control water (Appendix A, Table A-7) adjusted to a salinity of 2 ppt. Temperature was maintained at 20°C in an environmental chamber.

IV.1.6 Statistical Analysis

Lethal and sublethal effective concentrations were calculated using CETIS v. 1.1.2 (Tidepool Scientific Software, McKinleyville, CA, USA, 2006) following 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. Percentage minimum significant differences (PMSD) of Dunnett's multiple comparison procedure were calculated for all multiple concentration statistical tests.

IV.1.7 Analytical Chemistry

Water samples for analytical chemistry were collected upon preparation of test solutions using two acid-cleaned, 1-L amber-glass bottles. Samples for analysis of organic chemicals were immediately preserved by addition of 10 ml/l dichloromethylene. Samples were transported on ice and submitted to the California Department of Fish and Game Water Pollution Laboratory, Rancho Cordova, CA. Samples submitted for total and/or dissolved metals analysis were analyzed at the California Department of Fish and Game – Moss Landing Marine Laboratory, Moss Landing, CA.

For delta smelt tests, samples of all test solutions were submitted for chemical analysis. For fathead minnow tests, all test solutions prepared in "hatchery water" (HW) were submitted for whole water (dissolved and particulate) analysis, and the amounts of chemical detected in HW were used to calculate effect concentrations in both hatchery and synthetic laboratory control water. HW was found to contain 2.07 µg/L copper thus this amount was subtracted from measured copper concentrations of test concentrations for calculation of copper effect concentrations in synthetic laboratory control water.

For *H. azteca* tests with insecticides, all test solutions prepared in HW, as well as the highest, lowest and mid-range test concentrations prepared in synthetic laboratory control water were submitted for analysis. For the copper LC50 test, all test solutions prepared in synthetic laboratory control water, as well as the highest, lowest and mid-range test solutions prepared in HW were submitted for analysis. Were not all solutions were submitted, analytical data used to calculate effect concentrations were supplemented by data obtained for the corresponding test concentration in a different matrix.

For *C. dubia* chronic tests with insecticides, all test solutions in one matrix were submitted for analysis, and resulting data used to calculate effect concentrations in the second matrix. Samples from the *C. dubia* permethrin test were erroneously not submitted for analysis.

Organophosphate and pyrethroid insecticides were measured by gas chromatography (Agilent 6890 plus, Agilent Technologies Inc., Santa Clara, CA, USA) with dual columns (DB5 and DB17) and dual flame photometric detectors in phosphorous mode (OP insecticides), or dual micro-electron capture detectors (pyrethroid insecticides). Pyrethroids were confirmed using GC-MS or GC-MSMS.

IV.2 Results

IV.2.1 *H. transpacificus*

Toxic effect concentrations (96 h) for larval delta smelt were determined for ammonia/ium, un-ionized ammonia, copper (total and dissolved), the organophosphate

insecticide chlorpyrifos, and the pyrethroid insecticides esfenvalerate, bifenthrin, cyfluthrin and permethrin. These test were performed with treated Delta water obtained from the UCD FCCL adjusted to 900 µS/cm and pH 7.9. Results are presented both as measured and nominal concentrations (Tables 22 a, b). Among pesticides, delta smelt were most sensitive to esfenvalerate and bifenthrin, followed in order of decreasing sensitivity by cyfluthrin, permethrin and chlorpyrifos.

IV.2.2 *P. promelas*

Acute (96 h, 7 d) and chronic (7-d growth) effect concentrations for larval fathead minnow were determined for ammonia/ium, un-ionized ammonia, copper (total and dissolved), the organophosphate insecticide chlorpyrifos, and the pyrethroid insecticides esfenvalerate, bifenthrin, cyfluthrin and permethrin. Results are presented both as measured and nominal concentrations (Tables 23 a, b). These tests compared performance in hatchery water obtained from the UCD FCCL to performance in laboratory control water. Both types of water were adjusted to 900 µS/cm and pH 7.9.

As for delta smelt, the most toxic chemical was esfenvalerate, followed by cyfluthrin and bifenthrin. Permethrin was the least toxic pyrethroid. Copper was more toxic to larval fathead minnows than chlorpyrifos.

Water type did not affect acute toxicity of ammonia/um, chlorpyrifos, esfenvalerate or cyfluthrin. However, toxicity of bifenthrin was greater in "hatchery water (HW)" than in laboratory control water. Toxicity of permethrin was somewhat lower in hatchery water than in laboratory control water. Toxicity of copper was significantly lower in hatchery water than in control water, suggesting that dissolved organic carbon in Delta water rendered the metal less bioavailable. We were able to determine chronic effect concentrations for copper (7-d growth: 53 ug/L dissolved Cu^{+} in laboratory control water, and 101.9 µg/L dissolved Cu^{+} in hatchery water), chlorpyrifos and esfenvalerate.

Table 22 a. Measured acute effect concentrations of selected chemicals for larval delta smelt. Tests were performed using treated water from the delta smelt hatchery adjusted to EC 900 µS/cm.

Chemical	Test Date	Fish Age			96-h LC10		96-h LC50	
		[dph]	NOEC	LOEC	Estimate	95% C.I.	Estimate	95% C.I.
Ammonia Nitrogen (mg/L)	7/8/2009	47	14.4	29.0	5.38	< 1.9 - 9.38	11.8	8.1 - 18.5
Unionized Ammonia (mg/L)	7/8/2009	47	0.191	0.333	0.084	< 0.002 - 0.127	0.164	0.12 - 0.24
Copper (total) (ug/L)	5/14/2008	49	40.4	78.2	49.7	39.9 - 82.3	97.3	76.7 - 113.3
Copper (dissolved) (µg/L)	5/14/2008	49	41.4	76.2	50.1	40.9 - 80.1	92.1	74.7 - 110.8
Chlorpyrifos (µg/L)	5/5/2010	44	14.8	25.2	12.8	7.7 - 17.9	27.7	19.0 - 51.2
Esfenvalerate (µg/L)	4/7/2010	43	0.051	0.135	0.054	0 - 0.058	0.117	0.088 - 0.159
Bifenthrin (µg/L)	5/14/2008	49	0.120	0.260	0.095	0.061 - 0.117	0.143	0.12 - 0.17
Cyfluthrin (µg/L)	5/14/2008	49	0.407	0.890	0.26	0.067 - 0.357	0.42	0.26 - 0.56
Permethrin (µg/L)	4/7/2010	43	2.557	4.84	2.373	1.783 - 2.825	4.065	3.427 - 5.497

Table 22 b. Nominal acute effect concentrations of selected chemicals for larval delta smelt. Tests were performed using treated water from the delta smelt hatchery adjusted to EC 900 µS/cm.

Chemical	Test Date	Fish Age			96-h LC10		96-h LC50	
		[dph]	NOEC	LOEC	Estimate	95% C.I.	Estimate	95% C.I.
Ammonia Nitrogen (mg/L)	7/8/2009	47	20	40	7.44	< 2.5 - 13.25	16.45	11.35 - 25.57
Copper (total) (ug/L)	5/14/2008	49	37.5	75	45	19 - 55	85	76 - 95
Chlorpyrifos (µg/L)	5/5/2010	44	25	50	13.8	< 6.25 - 38.8	54.7	34.5 - 86.4
Esfenvalerate (µg/L)	4/7/2010	43	0.150	0.300	0.165	0.144 - 0.180	0.280	0.228 - 0.366
Bifenthrin (µg/L)	5/14/2008	49	0.250	0.500	0.215	0.140 - 0.262	0.305	0.246 - 0.359
Cyfluthrin (µg/L)	5/14/2008	49	0.500	1.000	0.160	0.129 - 0.466	0.454	0.393 - 0.523
Permethrin (µg/L)	4/7/2010	43	5	10	4.695	3.127 - 5.417	8.295	6.658 - 11.220

Table 23 a. Measured acute and chronic effect concentrations of selected chemicals for larval (<48 h old) fathead minnow. Tests were performed using both laboratory control water (DIEPAMH) and water from the delta smelt hatchery (HW) adjusted to EC 900 µS/cm. The 96-h ECs are calculated from survival recorded on day 4.

Chemical	Matrix	Test Date	96-h Survival		96-h LC10		96-h LC50		7-d Survival		7-d LC10		7-d LC50	
			NOEC	LOEC	Estimate	95% C.I.	Estimate	95% C.I.	NOEC	LOEC	Estimate	95% C.I.	Estimate	95% C.I.
Total Ammonia/um	DIEPAMHR	9/17/2008	15	30.8	17.1	16 - 21	28.9	22-45	15	30.8	17.1	16-21	28.9	22-45
(mg/L)	HW		15.2	29.8	16.0	15 - 16	20.9	20-21	15.2	29.8	16.0	15-16	20.9	20-21
Un-ionized Ammonia	DIEPAMHR	9/17/2008	0.518	1.004	0.60	0.56-0.73	0.95	0.75 - 1.54	0.518	1.004	0.60	0.56 - 0.73	0.95	0.75 - 1.54
(mg/L)	HW		0.629	1.121	0.66	0.63-0.68	0.85	0.83 - 0.86	0.629	1.121	0.66	0.63 - 0.68	0.85	0.83 - 0.86
Copper	DIEPAMHR	8/7/2008	32.2	67.1	49	45-63	103	92-122	32.2	67.1	40	36-50	85	67-103
(total; µg/L)	HW		132	260	132	81-150	216	188-248	69.2	132	90	79-117	162	146-180
Copper	DIEPAMHR	8/7/2008	31.7	60.6	46	43 - 58	96	85 - 115	31.7	60.6	39	35 - 47	78	60 - 96
(dissolved; µg/L)	HW		125	238	125	74-141	200	175-228	62.3	125	82	72-109	151	136-168
Chlorpyrifos (µg/L)	DIEPAMHR	8/19/2008	144	311	170	128 - 204	> 311	NA	144	311	141	90 - 165	252	202 - 347
	HW		144	311	159	137 - 180	> 311	NA	144	311	100	48 - 170	222	172 - 299
Esfenvalerate (µg/L)	DIEPAMHR	8/19/2008	0.500	0.920	0.536	0.52 - 0.55	0.74	0.72 - 0.76	0.500	0.920	0.53	0.52 - 0.54	0.69	0.67 - 0.71
	HW		0.500	0.920	0.516	0.49 - 0.54	0.67	0.65 - 0.68	0.500	0.920	0.52	0.49 - 0.53	0.67	0.65 - 0.68
Bifenthrin (µg/L)	DIEPAMHR	9/24/2008	0.7	1.4	0.70	0.45-0.84	1.19	0.94 - 1.53	0.7	1.4	0.54	0.43 - 0.82	1.00	0.87 - 1.12
	HW		0.4	0.7	0.42	0.39-0.53	0.92	0.43 - 1.50	0.4	0.7	0.42	0.39 - 0.48	0.76	0.50 - 1.03
Cyfluthrin (µg/L)	DIEPAMHR	7/7/2009	0.645	1.11	0.67	0.64-0.69	0.85	0.83 - 0.88	0.645	1.11	0.56	0.25 - 0.74	0.82	0.78 - 0.85
	HW		0.645	1.11	0.66	0.63-0.69	0.83	0.81 - 0.85	0.645	1.11	0.67	0.64 - 0.69	0.84	0.82 - 0.85
Permethrin (µg/L)	DIEPAMHR	7/7/2009	0.49	1.20	0.72	0.58-1.06	1.55	1.22 - 1.74	0.49	1.20	0.64	0.51 - 0.81	1.43	0.85 - 1.70
	HW		1.20	2.52	1.73	1.63 - 1.81			1.20	2.52	1.22	0.68 - 1.34	1.72	1.61 - 1.81

Table 23b. Measured chronic effect concentrations of selected chemicals for larval (<48 h old) fathead minnow. Tests were performed using both laboratory control water (DIEPAMH) and water from the delta smelt hatchery (HW) adjusted to EC 900 µS/cm.

Chemical	Matrix	Test Date	7-d Weight		7-d Weight EC10		7-d Weight EC25	
			NOEC	LOEC	Estimate	95% C.I.	Estimate	95% C.I.
Total Ammonia/um	DIEPAMHR	9/17/2008	30.8	> 30.8	> 30.8	NA	> 30.8	NA
(mg/L)	HW		15.2	> 15.2	> 15.2	NA	> 15.2	NA
Un-ionized Ammonia	DIEPAMHR	9/17/2008	1.004	> 1.004	> 1.004	NA	> 1.004	NA
(mg/L)	HW		0.629	> 0.629	> 0.629	NA	> 0.629	NA
Copper	DIEPAMHR	8/7/2008	67.1	129.9	41	12-45	58	41-75
(total; µg/L)	HW		69.2	132	72.14	<17.6-100.7	109.2	78.1- 82
Copper	DIEPAMHR	8/7/2008	60.6	123.3	39	12 - 42	53	40 - 67
(dissolved; µg/L)	HW		62.3	125	62.2	<1.72-238	101.9	70.9-171.4
Chlorpyrifos (µg/L)	DIEPAMHR	8/19/2008	43.2	82.4	7.8	0.5 - 67.4	66	35 - 106
	HW		43.2	82.4	28	<21.4-70.2	144	91 - 267
Esfenvalerate (µg/L)	DIEPAMHR	8/19/2008	0.920	> 0.920	> 0.920	-	> 0.920	-
	HW		0.500	> 0.500	> 0.50	-	> 0.50	-
Bifenthrin (µg/L)	DIEPAMHR	9/24/2008	1.4	>1.4	> 1.4	-	> 1.4	-
	HW		1.4	>1.4	> 1.4	-	> 1.4	-
Cyfluthrin (µg/L)	DIEPAMHR	7/7/2009	0.645	> 0.645	> 0.645	-	> 0.645	-
	HW		0.645	> 0.645	> 0.645	-	> 0.645	-
Permethrin (µg/L)	DIEPAMHR	7/7/2009	1.20	> 1.20	> 1.20	-	> 1.20	-
	HW		1.20	> 1.20	> 1.20	-	> 1.20	-

Table 23 c. Nominal acute and chronic effect concentrations of selected chemicals for larval fathead minnow. Tests were performed using both laboratory control water (DIEPAMH) and water from the delta smelt hatchery (HW) adjusted to EC 900 µS/cm. The 96-h ECs are calculated from survival recorded on day 4.

Chemical	Matrix	Test Date	96-h LC10		96-h LC50		7-d LC10		7-d LC50		7-d Weight EC10		7-d Weight EC25	
			Estimate	95% C.I.	Estimate	95% C.I.	Estimate	95% C.I.	Estimate	95% C.I.	Estimate	95% C.I.	Estimate	95% C.I.
Total Ammonia/um	DIEPAMHR	9/17/2008	22.7	21 - 28	37.6	28 - 57	22.7	21 - 28	37.6	28 - 57	> 40	NA	> 40	NA
(mg/L)	HW		21.1	20 - 22	27.8	27 - 29	21.1	20 - 22	27.8	27 - 29	> 20	NA	> 20	NA
Copper	DIEPAMHR	8/19/2008	47	43 - 66	99	87 - 113	38.9	35 - 48	80.1	70 - 91	39.7	16 - 44	56.6	48 - 76
(total; µg/L)	HW		125	74 - 142	207	180 - 239	83	72 - 109	154	138 - 172	62.36	< 15.6 - 93.4	102	71.1 - 174
Chlorpyrifos (µg/L)	DIEPAMHR	8/19/2008	233	180 - 272	>400	NA	196	109 - 227	331.0	272 - 442	8.8	0.4 - 78.4	79.1	39.5 - 133.4
	HW		219	189 - 244	>400	NA	128	53 -253	295.4	258 - 338	32.6	< 25 - 82.8	85.7	32 - 151
Esfenvalerate (µg/L)	DIEPAMHR	9/24/2008	0.541	0.52 - 0.55	0.779	0.76 - 0.81	0.54	0.52 - 0.54	0.72	0.70 - 0.75	>1.0	-	>1.0	-
	HW		0.519	489 - 537	0.694	0.67 - 0.71	0.52	0.49 - 0.53	0.69	0.67 - 0.71	> 0.500	-	> 0.500	-
Bifenthrin (µg/L)	DIEPAMHR	7/7/2009	1.25	0.71 - 1.49	2.10	1.70 - 3.02	0.91	0.63 - 1.45	1.76	1.58 - 2.04	> 2.5	-	> 2.5	-
	HW		0.72	0.57 - 1.16	1.66	0.73 - 2.70	0.69	0.58 - 0.91	1.39	0.84 - 1.83	> 1.25	-	> 1.25	-
Cyfluthrin (µg/L)	DIEPAMHR	7/7/2009	1.06	1.00 - 1.09	1.41	1.37 - 1.48	0.92	0.59 - 1.14	1.35	1.27 - 1.43	> 1.0	-	> 1.0	-
	HW		1.04	0.98 - 1.09	1.39	1.35 - 1.43	1.05	0.99 - 1.09	1.40	1.36 - 1.42	> 1.0	-	> 1.0	-
Permethrin (µg/L)	DIEPAMHR	9/17/2008	5.2	4.5 - 7.3	10.0	8.2 - 11.2	4.8	4.1 - 5.8	9.3	6.0 - 10.9	> 8.0	-	> 8.0	-
	HW		8.2	4.2 - 8.8	11.1	10.3 - 11.5	8.0	4.3 - 8.7	10.9	10.3 - 11.5	> 8.0	-	> 8.0	-

IV.2.3 *H. azteca*

Acute (96 h, 10 d) and chronic (10-d growth) effect concentrations for the amphipod *H. azteca* were determined for ammonia/ium, un-ionized ammonia, copper (total and dissolved), the organophosphate insecticides chlorpyrifos and diazinon, and the pyrethroid insecticides bifenthrin, cyfluthrin and permethrin. Results are presented both as measured and nominal concentrations (Tables 24 a-d). These tests compared performance in hatchery water obtained from the UCD FCCL to performance in laboratory control water. Both types of water were adjusted to 900 µS/cm and pH 7.9.

For *H. azteca*, the most toxic chemical was cyfluthrin with a 10-d LC50 of 1.74 (DIEPAMHR) and 1.86 (HW) ng/L, closely followed by bifenthrin with a 10-d LC50 of 2.7 (DIEPAMHR) and 2.3 (HW) ng/L. Permethrin was the least toxic pyrethroid tested with a 10-d LC50 of 63.9 (DIEPAMHR) and >90 (HW) ng/L. Amphipods were relatively sensitive to chlorppyrifos (10-d LC50: 105/103 ng/L), but far less sensitive to diazinon (10-d LC50: 3080/4310 ng/L) than other invertebrate test species. *H. azteca* is not sensitive to ammonia/um and copper compared to both fish and other invertebrate test species.

Water type did not affect the 10-d acute toxicity endpoint of ammonia/um, chlorpyrifos, bifenthrin or cyfluthrin. However, toxicity of diazinon and permethrin was somewhat lower in hatchery water than in laboratory control water. Toxicity of copper was significantly lower in hatchery water than in control water, suggesting that dissolved organic carbon in Delta water rendered the metal less bioavailable.

We determined chronic effect concentrations for ammonia/um (10-d growth: 20.2/52.5 mg/L NH_4^+; 0.688/1.52 mg/L NH_3), as well as for copper (10-d growth: 44/61 µg/L Cu^+), diazinon and bifenthrin. Effect concentrations were lower in laboratory control water than in hatchery water for ammonia/um, copper and diazinon, but higher for bifenthrin. For both organic pesticides, the chronic effect concentration was approximately half the 10-d LC50.

Table 24 a. Measured acute effect concentrations of selected chemicals for juvenile *Hyalella azteca*. Tests were performed using both laboratory control water (DIEPAMH) and water from the delta smelt hatchery (HW) adjusted to EC 900 µS/cm. The 96-h ECs are calculated from survival recorded on day 4.

Chemical	Date	Matrix	96-h LC10		96-h LC50		10-d LC10		10-d LC50	
			Estimate	95% C.I.	Estimate	95% C.I.	Estimate	95% C.I.	Estimate	95% C.I.
Total Ammonia/um (mg/L)	4/10/2009	DIEPAMHR	39.4	27.3 - 49.8	102.2	84-133	42.8	29.5 - 52.3	72.9	62 - 84
		HW	53.9	40.0 - 68.9	149.3	115 - 234	32.3	23.6 - 39.8	72.9	62 - 88
Un-ionized Ammonia (mg/L)	4/10/2009	DIEPAMHR	1.025	0.823 - 1.168	1.714	1.542 - 1.976	1.113	0.904 - 1.238	1.454	1.331 - 1.564
		HW	1.513	1.231 - 1.697	2.406	2.138 - 2.99	1.151	0.947 - 1.291	1.731	1.591 - 1.904
Copper (dissolved; µg/L)	4/10/2009	DIEPAMHR	271	244 - 303	449	377 - 554	122	36 - 133	178	165 - 197
		HW	303	230 - 627	613	474 - 737	199	152 - 287	343	312 - 369
Chlorpyrifos (ng/L)	1/15/2009	DIEPAMHR	77.9	65.4 - 117.6	153.6	95.7 - 183.3	71.1	65.6 - 75.9	105.2	88.2 - 141.7
		HW	78.3	75.4 - 83.4	146.6	131.4 - 161.8	72.1	70.5 - 73.8	102.6	91.2 - 114.7
Diazinon (ng/L)	12/30/2008	DIEPAMHR	2680	2460 - 3260	5900	4500 - 6950	1450	1300 - 1860	3080	2420 - 3670
		HW	3410	2980 - 5000	6737	4180 - 8210	3000	2790 - 3120	4310	4150 - 4460
Bifenthrin (ng/L)	1/14/2009	DIEPAMHR	2.4	2.2 - 2.6	3.9	3.4 - 4.3	2.1	2.1 - 2.2	2.7	2.5 - 3.0
		HW	2.9	1.4 - 3.4	4.3	4.0 - 4.9	1.2	1.0 - 1.6	2.3	1.6 - 4.5
Cyfluthrin (ng/L)	12/12/2008	DIEPAMHR	1.29	1.25 - 1.34	1.77	1.65 - 2.02	1.29	1.25 - 1.33	1.74	1.61 - 2.04
		HW	1.04	0.82 - 1.41	2.12	1.75 - 2.50	0.98	0.81 - 1.24	1.86	1.50 - 2.22
Permethrin (ng/L)	1/21/2009	DIEPAMHR	46.4	43.0 - 56.1	83.6	56.1 - > 90	43.1	40.0 - 45.0	63.9	59.6 - 68.1
		HW	> 90	-	> 90	-	44.1	1.6 - 56.0	>90	-

Table 24 b. Measured chronic effect concentrations of selected chemicals for juvenile *Hyalella azteca*. Tests were performed using both laboratory control water (DIEPAMH) and water from the delta smelt hatchery (HW) adjusted to EC 900 µS/cm.

Chemical	Date	Matrix	10-d Growth EC10		10-d Growth EC25	
			Estimate	95% C.I.	Estimate	95% C.I.
Total Ammonia/um	4/10/2009	DIEPAMHR	6.3	2.9 - 15.7	20.2	<4.85 - 28.8
(mg/L)		HW	40.5	<4.85 - 50.6	52.5	18 - 67
Un-ionized	4/10/2009	DIEPAMHR	0.292	0.180 - 0.587	0.688	0.107-0.876
Ammonia (mg/L)		HW	1.392	<0.279-1.501	1.516	0.954 - 1.64
Copper	4/10/2009	DIEPAMHR	17	15 - 22	44	37 - 57
(dissolved; µg/L)		HW	26	16 - 55	61	36 - 138
Chlorpyrifos	1/15/2009	DIEPAMHR	-	-	-	-
(ng/L)		HW	-	-	-	-
Diazinon (ng/L)	12/30/2008	DIEPAMHR	1030	< 410 - 1520	1410	< 410- 1990
		HW	930	590 - 1570	> 2800	-
Bifenthrin (ng/L)	1/14/2009	DIEPAMHR	0.5	< 0.6 - 1.2	1.3	< 0.6 - 2.3
		HW	0.2	0.1 - 0.2	0.5	0.4 - 0.7
Cyfluthrin (ng/L)	12/12/2008	DIEPAMHR	-	-	-	-
		HW	-	-	-	-
Permethrin (ng/L)	1/21/2009	DIEPAMHR	-	-	-	-
		HW	-	-	-	-

Table 24 c. Nominal acute effect concentrations of selected chemicals for juvenile *Hyalella azteca*. Tests were performed using both laboratory control water (DIEPAMH) and water from the delta smelt hatchery (HW) adjusted to EC 900 µS/cm. The 96-h ECs are calculated from survival recorded on day 4.

Chemical	Date	Matrix	96-h LC10		96-h LC50		10-d LC10		10-d LC50	
			Estimate	95% C.I.	Estimate	95% C.I.	Estimate	95% C.I.	Estimate	95% C.I.
Total Ammonia/um	4/10/2009	DIEPAMHR	52.3	36.6 - 65.5	130.9	108 - 169	57.3	40.5 - 69.1	94.3	81 - 108
(mg/L)		HW	69.5	46.1 - 88.9	192.9	149 - 304	41.6	30.3 - 51.4	94.3	80 - 114
Copper	4/10/2009	DIEPAMHR	291	224 - 343	484	422 - 553	125	36 - 140	174	165 - 183
(µg/L)		HW	352	274 - 412	570	500 - 650	207	153 - 295	318	293 - 344
Chlorpyrifos	1/15/2009	DIEPAMHR	75.2	61.4 - 158	152.7	91.2-185.8	67.6	62.1 - 72.6	102.4	84.3 - 145.6
(ng/L)		HW	138.8	124.2-154.1	138.8	124.2-154.1	96.7	86.1 - 108.0	101.9	93.2 - 111.4
Diazinon	12/30/2008	DIEPAMHR	2210	1410 - 2690	4440	3300 - 5470	1340	1150 - 2350	2670	2190 - 3080
(ng/L)		HW	2410	2000 - 3480	4900.00	2790 - 5810	2110	1950 - 2240	3120	3000 - 3270
Bifenthrin	1/14/2009	DIEPAMHR	5.38	4.56 - 6.27	9.01	9.01-11.35	4.48	4.28 - 4.59	6.88	6.09 - 8.00
(ng/L)		HW	7.87	5.29 - 8.69	11.48	10.57-13.28	4.36	3.98 - 4.99	6.73	5.31 - 11.75
Cyfluthrin	12/12/2008	DIEPAMHR	2.12	2.04 - 2.21	3.04	2.75 - 3.54	2.12	2.05 - 2.20	2.97	2.73 - 3.57
(ng/L)		HW	1.30	1.01 - 1.83	2.70	2.25 - 3.17	1.22	0.99 - 1.58	2.39	1.95 - 2.83
Permethrin	1/21/2009	DIEPAMHR	56.8	53.2 - 66.8	93.9	67.12- >100	53.24	50.0 - 55.3	74.6	70.3 - 78.7
(ng/L)		HW	> 100	-	>100	-	54.39	3.31 - 66.72	> 100	-

Table 24 d. Nominal chronic effect concentrations of selected chemicals for juvenile *Hyalella azteca*. Tests were performed using both laboratory control water (DIEPAMH) and water from the delta smelt hatchery (HW) adjusted to EC 900 µS/cm.

Chemical	Date	Matrix	10-d Growth EC10		10-d Growth EC25	
			Estimate	95% C.I.	Estimate	95% C.I.
Total Ammonia/um (mg/L)	4/10/2009	DIEPAMHR	8.5	3.7 - 21.3	26.7	0.2 - 38.4
		HW	51.7	< 6.25 - 65.3	67.9	22.9 - 87.5
Copper (µg/L)	4/10/2009	DIEPAMHR	18	15 - 24	45	38 - 62
		HW	24	13 - 57	60	32 - 147
Chlorpyrifos (ng/L)	1/15/2009	DIEPAMHR	-	-	-	-
		HW	-	-	-	-
Diazinon (ng/L)	12/30/2008	DIEPAMHR	930	< 500 - 1390	1270	0 - 1780
		HW	1050	550 - 2020	>2000	-
Bifenthrin (ng/L)	1/14/2009	DIEPAMHR	0.86	< 1.0 - 2.25	2.18	< 1.0 - 4.25
		HW	0.78	< 1.0 - 1.47	1.33	0.50 - 2.05
Cyfluthrin (ng/L)	12/12/2008	DIEPAMHR	-	-	-	-
		HW	0.29	0.16 - 0.66	0.88	0.45 - >3.9
Permethrin (ng/L)	1/21/2009	DIEPAMHR	1.30	0.50 - > 100	>50	-
		HW	16.11	13.63 - 37.16	23.42	15.38 - 79.28

IV.2.4 *C. dubia*

Two types of standard tests were performed to establish effect concentrations for the waterflea, *C. dubia*. The 96-h acute test follows different protocols and was expected to be more sensitive than the chronic test, where more food is provided for the organisms.

When comparing nominal effect concentrations from both testing methods, all chemicals tested were more toxic to *C. dubia* in the 96-h acute test than in the chronic test, with the possible exception of chlorpyrifos and diazinon (Tables 25 a-f). The 7-d LC50 derived from the chronic test was the same as the 96-h LC50 derived from the acute test for chlorpyrifos, and for diazinon, the 7-d LC50 (chronic test) was significantly lower than the 96-h LC50 (acute test). The differences were substantial for all pyrethroids tested. The acute test method resulted in 96-h LC50s that were significantly lower (>20-fold) than the 7-d LC50.

As opposed to *H. azteca* and both fish species, *C. dubia* was more sensitive to OP insecticides than to pyrethroids, and more sensitive than *H. azteca* to ammonia/um and copper (Table 28 a). Toxicity of copper was significantly lower in hatchery water than in control water, suggesting that dissolved organic carbon in Delta water rendered the metal less bioavailable (Table 25 d).

Table 25 a. Acute 96-h effect concentrations for *C. dubia*. Tests were performed using laboratory control water (SDEPAMH). All concentrations are nominal except ammonia/um concentrations.

Chemical	Test Date	96-h LC10		96-h LC50	
		Estimate	95% C.I.	Estimate	95% C.I.
Total Ammonia/um* (mg/L)	4/13/2010	5.8	<2.74 - 12.79	12.6	11 - 14
Un-ionized Ammonia* (mg/L)	4/13/2010	0.208	<0.147- .276	0.295	0.267-0.319
Copper (µg/L)	3/17/2010	19.7	13.4 - 20.4	26.3	24.5 - 26.8
Chlorpyrifos (ng/L)	3/11/2010	53.6	-	70.8	-
Diazinon (ng/L)	3/11/2010	207.4	195.5-218.5	277.8	268.9-286
Bifenthrin (ng/L)	3/2/2010	3.2	0.1 - 25.6	21.1	18.2 - 23.3
Cyfluthrin (ng/L)	3/2/2010	1.6	0.7 - 4.2	21.0	17.7 - 25.3
Permethrin (ng/L)	3/16/2010	278.5	256.2-311.3	428.7	331.3-651.3

* measured concentrations

Table 25 b. Nominal 96-h effect concentrations derived from chronic test with *C. dubia*. Tests were performed using laboratory control water (SDEPAMH). All concentrations are nominal except ammonia/um concentrations.

Chemical	Test Date	Matrix	96-h LC10		96-h LC50	
			Estimate	95% C.I.	Estimate	95% C.I.
Copper (dissolved; µg/L)	3/3/2010	SD900	37.5	2.4 - 43.1	59.6	49.5 - 84.2
		HW	94.5	84.2 - 150	182.9	133.7 - 204.2
Chlorpyrifos (ng/L)	10/8/2009	SD900	54.6	53.6 - 56.2	77.2	70.8 - 89.1
		HW	50.0	17.7 - 54.0	70.8	64.9 - 79.4
Diazinon (ng/L)	10/22/2009	SD900	268	-	353.6	-
		HW	256.3	11.1 - 268	345.0	329.9 - 353.6
Bifenthrin (ng/L)	3/18/2010	SS900	630	561.3 - 1072	1219	890.9 - 1414
		HW	561.3	540 - 630	890.9	734.9 - 1219
Cyfluthrin (ng/L)	4/7/2010	SD900	530.1	222 - 535.9	702.9	674.3 - 707.2
		HW	535.9	-	707.2	-
Permethrin (ng/L)	12/2/2009	SD900	1487	1380 - 2500	2806	2051 - 3402
	3/3/2010	HW	1487.0	1380 - 2500	2806.0	2051 - 3402

Table 25 c. Measured 96-h effect concentrations derived from chronic test with *C. dubia*. Tests were performed using laboratory control water (SDEPAMH).

Chemical	Test Date	Matrix	96-h LC10		96-h LC50	
			Estimate	95% C.I.	Estimate	95% C.I.
Copper (dissolved; µg/L)	3/3/2010	SD900	34.9	2.3 - 40.4	56.9	44.6 - 82.1
		HW	92.7	82.1 - 162	184.6	133.7 - 214.7
Chlorpyrifos (ng/L)	10/8/2009	SD900	20.9	20.7 - 21.2	24.7	23.7 - 26.5
		HW	20.0	1.7 - 20.8	23.7	22.7 - 25.0
Diazinon (ng/L)	10/22/2009	SD900	249.5	-	357.4	-
		HW	235.4	10.4 - 249.5	346.1	326.7 - 357.4
Bifenthrin (ng/L)	3/18/2010	SS900	329.7	308.1 - 432	506.2	403.8 - 552.7
		HW	308.1	301.3 - 329.7	403.8	360.8 - 506.2
Cyfluthrin (ng/L)	4/7/2010	SD900	543.9	229.4 - 549.5	708.2	681.3 - 712.3
		HW	549.5	-	712.3	-
Permethrin* (ng/L)	12/2/2009	SD900	NA	NA	NA	NA
	3/3/2010	HW	NA	NA	NA	NA

* concentrations not measured.

Table 25 d. Dissolved organic carbon (DOC) measurements in Hatchery Water and synthetic laboratory control water (SSEPAMH).

Water Type	Date	DOC (mg/L)
Filtered Hatchery Water	2/7/2010	6.19
SSEPAMH	2/8/2010	0.63
Filtered Hatchery Water	3/1/2010	7.61
SSEPAMH	3/1/2010	0.94
Filtered Hatchery Water	3/15/2010	7.25
SDEPAMH	3/18/2010	1.04
Filtered Hatchery Water	4/4/2010	6.29
SDEPAMH	4/2/2010	ND
Filtered Hatchery Water	5/3/2010	3.79

Table 25 e. Nominal acute and chronic effect concentrations for *C. dubia* 7-d survival and fecundity for selected chemicals. Tests were performed using both laboratory control water (SDEPAMH) and water from the delta smelt hatchery (HW) adjusted to EC 900 µS/cm.

Chemical	Date	Matrix	7-d LC10		7-d LC50		7-d Reproduction EC10		7-d Reproduction EC25	
			Estimate	95% C.I.	Estimate	95% C.I.	Estimate	95% C.I.	Estimate	95% C.I.
Copper (µg/L)	3/3/2010	SDEPAMH	37.5	2.4 - 40.9	53.1	47.3 - 59.5	2.20	1.1 - 11.8	17.1	5.3 - 40.9
		HW	106.1	86.2 - 160.8	194.6	150- 212.2	55.90	44.0 - 83.2	83.2	58.5 - 98.2
Chlorpyrifos (ng/L)	10/8/2009	SDEPAMH	53.6	-	70.8	-	52.4	35.1 - 54.2	59.3	54.5 - 61.3
		HW	50.0	17.7 - 53.6	68.1	64.9 - 70.8	17.6	9.0 - 51.4	53.8	21.5 - 58.5
Diazinon (ng/L)	10/22/2009	SDEPAMH	134.0	-	176.8	142-220.1	145.3	126.5-155.1	189.5	170.9-222.1
		HW	14.9	2.99-134	172.7	150.4-198.3	4.0	1.61-142.9	53.85	9.98-237.9
Bifenthrin (ng/L)	3/18/2010	SDEPAMH	500.0	315 - 535.9	680.5	609.6-707.2	298.4	15.2 - 344.4	388.9	298.2 - 520.4
		HW	280.6	270-315	445.5	367.5-609.6	290.7	78.1 - 315.3	364.6	310.7 - 446.6
Cyfluthrin (ng/L)	4/7/2010	SDEPAMH	511.7	177 - 536	689.2	610 - 707	91.3	3 - 203	171.8	25 - 282
		HW	535.9	-	707.2	-	9.8	4 - 96	140.0	49 - 177
Permethrin (ng/L)	12/2/2009	SDEPAMH	1250.0	787.5 - 2500	2806	1895 - 3402	209.6	12.25 - 725.7	899.8	498 - 1354
		HW	1250.0	787.5-2500	2806	1895 - 3402	251.1	9.7 - 771	815.3	357 - 1268

** poor test acceptability control (TAC) performance

Table 25 f. Measured acute and chronic effect concentrations for *C. dubia* 7-d survival and fecundity for selected chemicals. Tests were performed using both laboratory control water (SDEPAMH) and water from the delta smelt hatchery (HW) adjusted to EC 900 µS/cm.

Chemical	Date	Matrix	7-d LC10		7-d LC50		7-d Reproduction EC10		7-d Reproduction EC25	
			Estimate	95% C.I.	Estimate	95% C.I.	Estimate	95% C.I.	Estimate	95% C.I.
Copper	3/3/2010	SDEPAMH	34.9	2.3 - 38.8	50.4	43.1 - 59.0	2.4	1.1 - 21.2	20.7	5.1 - 39.4
(dissolved; µg/L)		HW	92.7	82.1 - 162	184.6	133.7 - 214.7	53.20	41.5 - 80.2	81.0	54.2 - 95.6
Chlorpyrifos (ng/L)	10/8/2009	SDEPAMH	20.7	-	23.7	-	20.6	18.4 - 20.8	21.9	21.2 - 22.1
		HW	20.0	1.7 - 20.7	23.2	22.4 - 23.7	7.7	1.52 - 20.8	21.3	9.13 - 22.1
Diazinon (ng/L)	10/22/2009	SDEPAMH	130.8	-	167.5	-	139.1	123.1-148.1	176.5	160.3 - 208
		HW	14.0	2.87-130.8	164.1	146.8- 85.7	44.1	2.41- 228	176.2	20.5 - 264.9
Bifenthrin (ng/L)	3/18/2010	SDEPAMH**	288.0	209.8 - 299.9	344.9	323.4 - 352.7	203.1	8.0 - 240	245.4	203 - 302
		HW	193.8	188.7 - 209.8	266.1	233.2 - 323.4	198.5	86.9 - 208.7	231.9	211.6 - 262.9
Cyfluthrin (ng/L)	4/7/2010	SDEPAMH	537.8	183 -550	703.8	620 -712	64.9	1.8 - 270.9	163.9	12.0 - 307.7
		HW	549.5	NA	712.3	NA	9.7	3.6 - 112.5	127.6	43.3 - 183.3
Permethrin (ng/L)	12/2/2009	SDEPAMH	*	*	*	*	*	*	*	*
		HW	*	*	*	*	*	*	*	*

* measured concentrations not available
** poor TAC performance

IV.2.5 *E. affinis*

Acute (96-h) effect concentrations were determined for ammonia, copper, the organophosphate insecticide chlorpyrifos, and the pyrethroid insecticides bifenthrin, cyfluthrin, and permethrin by Dr. Teh (UC Davis, CA). In addition, the UCD ATL performed several LC50 tests with ammonia and copper (900 µS/cm and 2 ppt salinity), as well as rangefinder tests for insecticides. Please refer to chapter III.5 (Teh et al.). Results obtained by UCD ATL are presented in Tables 26 a, b. below.

The sensitivity of *E. affinis* to copper was determined in multiple tests (Table). Tests conducted at a salinity of 2 ppt and a temperature of 20°C resulted in 48-h LC50s and 96-h LC50s of 210 - >300 µg/L, and 31.6 – 109.2 µg/L, respectively. At a conductivity of 900 µS/cm, the 96-h LC50 was 72.6 µg/L copper (dissolved). These results are in sharp contrast to 96-h LC values reported by Teh et al. (2009) of 3.48 µg/L, who tested under similar conditions (T=20°C, pH 8.0) and at a salinity of 2 ppt.

While UCD ATL results for copper are 10-33 times higher than those reported by Teh et al., LC50 values for total ammonia/um were similar (depending on pH). Effect concentrations for ammonia/um suggest, that *E. affinis* is far more sensitive to ammonia/um than *H. azteca*. Effect concentrations for the insecticides chlorpyrifos, cyfluthrin, permethrin and bifenthrin determined by both labs were similar, and demonstrate that *E. affinis* is less sensitive to pyrethroids, and chlorpyrifos than other common invertebrate test species.

Table 26 a. Nominal effect concentrations for copper in *E. affinis* reference toxicant tests conducted at a salinity of 2 ppt.

Date	Endpoint	Control Survival (%)	Copper (ug/L)					
			NOEC	LOEC	LC10		LC50	
					Estimate	95% C.I.	Estimate	95% C.I.
1/31/2010	24-hr Survival	100	256	>256	-	-	-	-
	48-hr Survival	100	128	256	8	0.6 - 64	210	128 - >256
	72-hr Survival	90	128	256	9.3	0.5 - 19.4	76	16 - 137
	96-hr Survival	90	64	128	1.9	0.4 - 16.8	70	15 - 83
2/26/2010	24-hr Survival	100	300	>300	>300	-	>300	-
	48-hr Survival	100	300	>300	189	60 - >300	>300	-
	72-hr Survival	100	300	>300	37.5	24 - 172	>300	-
	96-hr Survival	80	300	>300	39.9	3 - 70	71.8	53 - 246
3/8/2010*	24-hr Survival	100	300	>300	150	95 - 300	>300	-
	48-hr Survival	90	300	>300	110.4	90 - 300	>300	-
	72-hr Survival	90	150	300	86.5	83 - 110	156.7	123 - >300
	96-hr Survival	50	300	>300	67.6	2 - 84	109.2	53 - 162
3/26/10*	24-hr Survival	100	300	>300	75	27 - >300	>300	-
	48-hr Survival	80	300	>300	51	27 - 172	>300	-
	72-hr Survival	80	300	>300	35	22 - 43	63.1	50 - >300
	96-hr Survival	50	300	>300	3.4	0.5 - 40	31.6	5.7 - 56

*: Control survival was < 80%

Table 26 b. Acute effect concentrations (UCD-ATL) for *E. affinis* for selected chemicals. Tests were performed using laboratory control water (SSEPAMH) adjusted to EC 900 µS/cm and pH 7.9.

Chemical	Test Date	Notes	NOEC	LOEC	96-h LC50	
					Estimate	95% C.I.
Nominal Ammonia (mg/L)	3/9/2010	Nominal	6.3	12.5	2.5	1.3 - 6.9
Ammonia Nitrogen (mg/L)	3/9/2010	Measured	7.0	14.7	3.1	1.5 - 7.8
Un-ionized Ammonia (mg/L)	3/9/2010	Measured	0.222	0.434	0.117	0.076 - 0.250
Nominal Copper (ug/L)	3/3/2010	Nominal	150	300	75.0	30.2 - 172.3
Dissolved Copper (ug/L)	3/3/2010	Measured	151	305	72.6	27.1 - 174.9
Chlorpyrifos (ng/L)	3/12/2010	Rangefinder test, 4 rep./treatment	N/A*		1092	<25 - 1216
Diazinon (ng/L)	3/12/2010	Rangefinder test	N/A*		No Dose Response	
Bifenthrin (ng/L)	3/10/2010	Rangefinder test	N/A*		26.08	<2 - 243.6
Cyfluthrin (ng/L)	3/10/2010	Rangefinder test	N/A*		31.30	<2 - 115.5
Permethrin (ng/L)	3/12/2010	Rangefinder test	N/A*		467.8	<30 - 1731

* replication too low to derive value.

IV.2.6 Interspecies Comparison

IV.2.6.1 Fish

Comparison of effect concentrations derived from tests using hatchery water adjusted to 900 µS/cm and ph 7.9 showed that larval delta smelt (although older and larger) were more sensitive than fathead minnow to ammonia/um, copper, and all insecticides tested with the exception of permethrin (Table 27).

Table 27. Comparison of the NOEC, LOEC and 96-h LC50s of 39 – 51 day old delta smelt and <48 h old fathead minnows in hatchery water to selected chemicals. All effect concentrations are based on measured concentrations.

Chemical	Delta Smelt (DS)			Fathead Minnow (FHM)			Relative Sensitivity
	NOEC	LOEC	LC50	NOEC	LOEC	LC50	LC50 FHM/ LC50 DS
Ammonia Nitrogen* (mg/L)	14.4	29.0	11.8/11.6	15.2	29.8	20.9	**1.8**
Unionized Ammonia* (mg/L)	0.191	0.333	0.164/0.147	0.629	1.121	0.85	**5.2/5.8**
Copper (total; µg/L)	40.4	78.2	97.3	132	260	216	**2.2**
Copper (dissolved; µg/L)	41.4	76.2	92.1	125	238	200	**2.2**
Chlorpyrifos (µg/L)	14.8	25.2	27.7	144	311	>311	**>11.2**
Esfenvalerate (µg/L)	0.051	0.135	0.117	0.500	0.920	0.670	**6.4**
Bifenthrin (µg/L)	0.120	0.260	0.143	0.4	0.7	0.920	**6.4**
Cyfluthrin (µg/L)	0.407	0.890	0.420	0.645	1.11	0.830	**2**
Permethrin (µg/L)	2.557	4.84	4.065	1.20	2.52	1.730	**0.43**

* two tests were performed

IV.2.6.2 Invertebrates

Comparison of effect concentrations for *H. azteca, C. dubia* and *E. affinis* shows markedly different species sensitivities to ammonia/um, copper and pesticides (Tables 28 a, b). The copepod *E. affinis* and the waterflea *C. dubia* were the most sensitive species with regard to ammonia/um, while *H. azteca* was far less sensitive to this compound than all invertebrate and fish species tested. Similarly, *H. azteca* was least sensitive to copper, while *C. dubia* was most sensitive. Values for *E. affinis* varied widely. However, a series of four LC50 tests with copper conducted at 2 ppt (same salinity as used in tests conducted by Teh et al., 2009) resulted in nominal 96-h LC50 values of 31.6 – 109.2 µg/L (Table 26 a). At a conductivity of 900 µS/cm the 96-h LC50 was 75 µg/L. This suggests that the low LC50 of 3.48 µg/L copper must be considered an outlier.

C. dubia was most sensitive to chlorpyrifos, and 16-fold more sensitive to diazinon than *H. azteca,* while *E. affinis* was the least sensitive species with regard to chlorpyrifos. *H. azteca* was the most sensitive to all pyrethroid insecticides tested (bifenthrin, cyfluthrin and permethrin). *C. dubia* showed the least sensitivity to these materials, while the sensitivity of *E. affinis* was intermediate.

In order to investigate if standard toxicity endpoints are protective of copepod species, Table 28 b compares effect concentrations of *E. affinis* to toxicity endpoints of *H. azteca* and *C. dubia* tests considered to be most sensitive. *C. dubia* reproduction was most sensitive to copper and both organophosphate insecticides, while *H. azteca* 10-d survival and growth were the most sensitive endpoints for pyrethroid insecticides. The *C. dubia* chronic endpoint (reproduction) was far less sensitive than the acute endpoint (96-h survival) for pyrethroid toxicity, likely due to the differences in test protocols.

Table 28 a. Comparison of 96-h LC50 (nominal unless otherwise noted) of *H. azteca*, *E. affinis* and *C. dubia* exposed in laboratory control water to selected chemicals. Sensitivity ratios are derived using nominal concentrations, except for ammonia/um (*C. dubia/H. azteca* comparison).

Chemical	*H. azteca* 96-h LC50 [a] (measured)	*H. azteca* 96-h LC50 [a] (nominal)	*E. affinis* 96-h LC50 [a]	*E. affinis* 96-h LC50 [b]	*C.dubia* Acute 96-h LC50 [a]	*H. azteca: C. dubia*	*H. azteca: E. affinis*	*E. affinis: C. dubia*
Total Ammonia/um (mg/L)	102.2 [c]	130.9	3.1 [c]	10.97	12.6 [c]	**10.4**	**33/10.4**	**0.25/0.87**
Un-ionized Ammonia (mg/L)	1.714 [c]	4.36	0.117 [c]	0.78	0.42 [c]	**10.4**	**14.6/2.2**	**0.28/1.86**
Copper (µg/L)	449.0 [c]	484	75.0	3.48	26.3	**17.1**	**6.5/139**	**2.85/0.13**
Chlorpyrifos (µg/L)	0.153 [c]	0.153	1.092	0.803	0.071	**2.2**	**0.14/0.19**	**15.4/11.3**
Diazinon (µg/L)	5.90 [c]	4.44	-	-	0.278	**16.0**	**-**	**-**
Bifenthrin (µg/L)	0.0039 [c]	0.009	0.026 [d]	0.011*	0.021	**0.43**	**0.34/0.82**	**1.24/0.52**
Cyfluthrin (µg/L)	0.0018 [c]	0.003	0.031 [d]	0.013	0.021	**0.15**	**0.10/0.23**	**1.48/0.62**
Permethrin (µg/L)	0.0836 [c]	0.094	0.468 [d]	0.158**	0.429	**0.22**	**0.20/0.59**	**1.09/0.37**

[a] Data generated at UCD ATL; experimental conditions: SC= 900 µS/cm, pH 7.9, T= 20.0ºC

[b] Data obtained from S. Teh (UC Davis); experimental conditions: salinity=2 ppt, pH 8.1, T= 20.0ºC

[c] measured or calculated from measured concentrations

[d] derived from rangefinder tests

* measured concentrations ranged from 2 - 18 ng/L (nominal : 4 – 32 ng/L)

** measured concentrations did not follow a concentration gradient

Table 28 b. Comparison of *E. affinis* 96-h LC50s to effect concentrations using the most sensitive endpoints for *H. azteca (10-d survival, growth)*, and *C. dubia* (7-d fecundity) exposed in laboratory control water to selected chemicals. Concentrations were nominal unless otherwise noted.

Chemical	*E. affinis* 96-h LC50 [a]	*E. affinis* 96-h LC50 [b]	*H. azteca* 10-d LC50 [a]	*H. azteca* 10-d Growth EC25 [a]	*C.dubia* Acute 96-h LC50 [a]	*C.dubia* Chronic, 7-d fecundity EC25 [a]
Total Ammonia/um (mg/L)	3.1 [c]	10.97	72.9 [c]	20.2 [c]	12.6 [c]	-
Un-ionized Ammonia (mg/L)	0.117 [c]	0.78	1.45 [c]	0.688 [c]	0.42 [c]	-
Copper (µg/L)	75.0	3.48	178.0 [c]	44 [c]	26.3	20.7 [c]
Chlorpyrifos (µg/L)	1.092	0.803	0.105 [c]	-	0.071	0.022 [c]
Diazinon (µg/L)	-	-	3.08 [c]	1.41 [c]	0.278	0.177 [c]
Bifenthrin (µg/L)	0.026 [d]	0.011	0.0027 [c]	0.0013 [c]	0.021	0.245 [c]
Cyfluthrin (µg/L)	0.031 [d]	0.013	0.0018 [c]	-	0.021	0.164 [c]
Permethrin (µg/L)	0.468 [d]	0.158	0.0640 [c]	<0.050 [c]	0.429	0.900

[a] Data generated at UCD ATL; experimental conditions: SC= 900 µS/cm, pH 7.9, T= 20.0ºC

[b] Data obtained from S. Teh (UC Davis); experimental conditions: salinity=2 ppt, pH 8.1, T= 20.0ºC

[c] measured or calculated from measured concentrations

[d] derived from rangefinder test

IV.3 Acute Toxicity of Ammonia, Copper, and Pesticides to *Eurytemora affinis*, of the San Francisco Estuary

Final Report
Submitted to:

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Executive Summary

Toxicity testing (96-h) of ambient surface waters in April-May 2008 from several locations in the North and South Delta-San Francisco Estuary (SFE) was shown to significantly affect the survival of *Eurytemora affinis.* Although chemical contaminants such as ammonia, bifenthrin, copper diuron, lambda cyhalothrin, and polyaromatic hydrocarbons have been detected in ambient waters, the impacts of these contaminants to pelagic organisms in the SFE food web are critically unknown particularly to the dominant zooplankton, i.e., *E. affinis*. The acute toxicity of ammonia, bifenthrin, chlorpyrifos, copper, cyfluthrin and permethrin to *E. affinis* was addressed in the current study as shown by the results of 96hr-LC50 values of the different contaminants: 1) ammonia - 10.97 mg/L total ammonia or 0.78 mg/L unionized ammonia at pH 8.1, 7.56 mg/L total ammonia or 0.12 mg/L unionized ammonia at pH7.6, and 10.93 mg/L total ammonia or 0.068 mg/L unionized ammonia at pH7.2; 2) bifenthrin - 11.37 ng/L, 3) chlorpyrifos - 803.20 ng/L 4) copper - 3.48 μg/L, 5) cyfluthrin - 12.72 ng/L and 6) permethrin -158.08 ng/L. Current findings indicated that *E. affinis* were sensitive to ammonia, copper, and pyrethroid pesticides (bifenthrin, cyfluthrin, and permethrin) and organophosphate insecticide (chlorpyrifos). Based on the results of this study, it is likely that the toxicities observed in *E. affinis* in 2008 may have been due, in part, to the presence of some of these chemicals in examined ambient waters. The potential impact of one or additive effects of these chemicals pose serious implications to the health and survival of zooplankton as important components of the SFE food web.

Introduction

Eurytemora affinis is an important food source to higher trophic level pelagic fish such as delta smelt, threadfin shad, and longfin smelt in the San Francisco Estuary (SFE). Previous study in this laboratory revealed that ambient surface waters from several locations in the North and South Delta in April-May 2008 showed significant effects to *E. affinis* survival (Teh *et al*., 2008). The initial detection of several chemical contaminants including ammonia, bifenthrin, chlorpyrifos, copper, cyfluthrin and permethrin in ambient waters prompted the need to examine their acute toxicity to *E. affinis*. Assessing the 96-hour LC50 values to establish the toxicity of these contaminants to *E. affinis* under controlled laboratory conditions was the main objective of the current study.

Experimental Details

1. Copepods

Brood stock of *E. affinis* was grown in aerated 120 L tanks placed in an environmentally controlled room at 20 ± 1 ^{o}C. Water quality in the tank including dissolved oxygen (>8 mg/L), pH (8.0 ± 0.1), water hardness (100 mg/L), salinity (2.0 ppt), and ammonia (<1

µg/L) were monitored weekly. An equal biovolume of the Instant Algae (*Nannochloropsis* and *Pavlova*) mix was given as food at 400 µg $C.L^{-1}$.day-1.

2. *Chemicals*

Stock solutions of ammonium chloride (10.0 g/L), bifenthrin (8.0 mg/L), chlorpyrifos (4.0 mg/L) copper chloride (4.0 mg/L), cyfluthrin (4.0 mg/L), and permethrin (8.0 mg/L) were prepared by personnel of Aquatic Toxicology Laboratory at UC Davis. The concentrations of the chemical used were: 1) bifenthrin (methanol control, 4.0, 8.0, 16.0, 32.0, and 64.0 ng/L), 2) chlorpyrifos (methanol control, 300, 600, 900, 1200, 1500 ng/L), 3) cyfluthrin (methanol control, 1.0, 3.0, 5.0, 7.0, 9.0 ng/L), and 4) permethrin (methanol control, 150, 175, 200, 225, 250 ng/L). Methanol was used as solvent for these chemicals, and therefore served as control using the highest concentration in each of the chemical treatments. The concentrations used for ammonia were: 1) 0.0, 10.0, 15.0, 20.0, 25.0, and 30.0 mg/L at pH 8.1, 2) 0.0, 10.0, 15.0, 20.0, 25.0, and 30.0 mg/L at pH7.6, and 3) 0.0, 4.0, 6.0, 8.0, 10.0, and 12.0 mg/L at pH7.2 that were prepared by diluting the ammonium chloride stock solution with culture water and the pH adjusted with 1N HCl. The concentrations used for copper chloride were 0.0, 1.0, 2.0, 4.0, 6.0, and 8.0 µg/L. Graded concentrations of these chemicals were prepared by diluting the stock solution with culture water (same source of water as used for culturing the *E. affinis*) 30-45 minutes prior to the initiation of the 96-hour exposures.

3. *Acute Toxicity Test*

Groups of juvenile *E. affinis* (N = 20 per replicate; three replicates per concentration) were exposed separately to ammonia, bifenthrin, chlorpyrifos, copper, cyfluthrin and permethrin using the standard static renewal method for acute toxicity testing (1993). The test conditions used for the acute toxicity tests for ammonia, bifenthrin, chlorpyrifos, copper, cyfluthrin and permethrin are shown in Table 1. Briefly, Copepods were fed with nutritious algae and 80% of the tested water was replaced at 24, 48, and 72 h with newly prepared corresponding treatment solutions previously acclimated to 20 ^{o}C. Mortalities were recorded daily for 4 days. At the end of 96 hr, the number of survivors in each beaker was counted to derive the mean percentage survival of *E. affinis* exposed to each chemical concentration. The estimated 96-hour LC50 values (Lethal Concentration causing 50% mortality of the *E. affinis*) were calculated using the U.S. Environmental Protection Agency Probit Analysis Program v1.5 (http://www.epa.gov/nerleerd/stat2.htm).

4. *Water parameters and chemical analysis*

Water quality was monitored and recorded daily for each of the acute toxicity trials. Unionized ammonia was calculated from total ammonia nitrogen using free ammonia calculator (http://cobweb.ecn.purdue.edu/~piwc/w3-research/free-ammonia/nh3.html). The concentrations of the chemicals used for the toxicity trials will be verified at the Aquatic Toxicology Laboratory at UC Davis by testing I L subsamples of each of the chemical concentrations prior to the exposure trials.

Results and Discussions

The mean survival (%) of *E. affinis* at the end of 96 hour of toxicity testing is given in Table 2. The 96hr-LC10 and 96hr-LC50 values with 95% confidence intervals as calculated using the USEPA Probit Analysis Program v1.5 are shown in Table 3.
The data demonstrates that juvenile *E. affinis* are sensitive to the ammonia, copper, pyrethroid pesticides (.bifenthrin, cyfluthrin and permethrin), and organophosphate insecticide (chlorpyrifos). This pilot study aimed to establish LC50 values for *E. affinis* to support the hypothesis that ambient water samples from certain locations in the SFE are toxic to *E. affinis.* Based on the results of this study, it is likely that the toxicities observed in *E. affinis* in 2008 may have been due, in part, to the presence of these chemicals in examined ambient waters. The potential impact of one or additive effects of these chemicals pose serious implications to the health and survival of zooplankton as important components of the SFE food web.

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Table 1 Test conditions used for *Eurytemora affinis*

Temperature (°C)	**20 ± 0.1**
Salinity (ppt)	**2**
pH	**8.0 ± 0.1**
Conductivity (µmhos)	**3000**
Hardness (mg/L)	**360**
Alkalinity (mg/L)	**60**
Acceptability in control survival	**≥80%**
Size of test beaker (mL)	**600**
Volume of test solution (mL)	**500**
Life stage of copepods	**Juvenile**
# of copepods	**20**
# of replicates per concentration	**3**
# of concentrations	**6**
Feeding regime	**Daily**
Static-renewal test Duration	**24-96 h**

Table 2 Mean % survivorship of *E. affinis* at the end of 96 hour exposure

Chemicals	Concentration	% Survivorship
Ammonia mg/L at pH 8.1	Control	96.66
	10	56.66
	15	20.00
	20	5.00
	25	0
	30	0
Ammonia mg/L at pH 7.6	Control	88.33
	10	16.66
	15	0
	20	0
	25	0
	30	0
Ammonia mg/L at pH 7.2	Control	88.33
	4	60.00
	6	56.66
	8	55.00
	10	46.66
	12	35.00
Bifenthrin ng/L (pptr)	Methanol control	85.00
	4	75.00
	8	43.33
	16	38.33
	32	16.67
	64	3.33
Chlorpyrifos ng/L (pptr)	Methanol control	83.33
	300	76.66
	600	65.00
	900	26.66
	1200	18.33
	1500	15.00
Copper µg/L (ppb)	Control	88.33
	1	88.33
	2	61.66
	4	23.33
	6	30.00
	8	13.33

Cyfluthrin ng/L (pptr)	Methanol control	88.33
	1	85.00
	3	68.33
	5	56.66
	7	68.33
	9	46.66
Permethrin ng/L (pptr)	methanol Control	88.33
	150	46.66
	175	35
	200	31.66
	225	25
	250	11.66

Table 3 Estimates LC 10 and 50 values of *E. affinis* calculated using Probit Analysis (95% confidence intervals are indicated in parentheses)

Chemicals	96hr-LC10	96hr-LC50
Total Ammonia (mg/L; pH8.1)	7.01 (5.50, 8.71)	10.97 (9.76, 11.96)
Unionized Ammonia (mg/L; pH8.1)	0.46 (0.35, 0.55)	0.78 (0.68, 0.86)
Total Ammonia (mg/L; pH7.6)	5.02 (1.42, 6.85)	7.56 (4.07, 8.95)
Unionized Ammonia (mg/L; pH7.6)	0.08 (0.02, 0.11)	0.12 (0.06, 0.14)
Total Ammonia (mg/L; pH7.2)	1.82 (0, 2.79)	10.93 (7.34,49.0)
Unionized Ammonia (mg/L; pH7.2)	0.011 (0.0, 0.017)	0.068 (0.046, 0.306)
Bifenthrin (ng/L; pptr)	2.76 (1.27, 4.43)	11.37 (8.04, 14.80)
Chlorpyrifos (ng/L; pptr)	384.49 (211.81, 515.58)	803.20 (640.17, 926.41)
Copper (µg/L; ppb)	1.42 (0.61, 1.45)	3.48 (2.85, 4.15)
Cyfluthrin (ng/L; pptr)	1.40 (0.05, 2.89)	12.72 (8.05, 55.55)
Permethrin (ng/L; pptr)	83.37 (38.71, 110.83)	158.08 (125.55, 175.99

Manuscript *in preparation*.

Evaluating the Suitability of *Hyalella azteca* for Chronic Water Column Toxicity Testing.

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Abstract

Standard US EPA laboratory tests are used to monitor toxicity in surface waters. The waterflea, *Ceriodaphnia dubia,* is considered one of the most sensitive test organism for detecting insecticide toxicity in freshwater environments. However, several inland water bodies within California typically have conductivities higher than this species can tolerate. The euryhaline amphipod, *Hyalella azteca,* has the potential to be a valuable surrogate test species under such conditions. Before tests with *H. azteca* can effectively replace or complement *C. dubia* tests in monitoring programs, information must be generated on the sensitivity of the species to toxicants, the method's ability to detect statistical differences, and the availability of protocols for Toxicity Identification Evaluation (TIE) procedures.

This study compares the sensitivity of two water column toxicity tests, the chronic US EPA *C. dubia* test, and a 10-d *H. azteca* test. Organism sensitivity was evaluated through the collection of concentration effect data for four current-use insecticides. Test sensitivity was evaluated by comparing the percent minimum significant differences (PMSDs). Factors affecting the ability to identify pyrethroids pesticides in TIE procedures were addressed, including adsorption to container walls, selecting effective piperonyl buteoxide (PBO) concentrations and quantifying synergism between PBO and pyrethroids.

H. azteca was more sensitive to pyrethroids and less sensitive to organophosphate (OP) pesticides than *C. dubia*. The *H. azteca* survival endpoint was the most sensitive, followed by the *C. dubia* reproduction endpoint. Adsorption of insecticides to sampling containers was greatest within the first 72 h of sample storage with approximately 80% loss for all insecticides studied. Fifty and 100 µg/L piperonyl-butoxide (PBO) accelerated mortality in conjunction with one toxic unit of two pyrethroids, bifenthrin or permethrin, and delayed mortality in the presence of the organophosphate, chlorpyrifos. Piperonyl-butoxide at a concentration of 25 µg/L synergized pyrethroid toxicity up to 9-fold. Our results show that *H. azteca* is a viable test species for evaluating ecosystem health, especially when the conductivity range of samples falls between 200 and 10,000 µS/cm, and pyrethroids may be present in the sample

Key Words: *Hyalella azteca*, *Ceriodaphnia dubia*, insecticides, organophosphates, pyrethroids, piperonyl butoxide

Introduction

Ceriodaphnia dubia has been used as a surrogate species in standard US EPA toxicity tests since the 1980s for ambient monitoring in California. Data gathered from these tests have been used by regulators to provide weight of evidence for TMDL listings, meeting Basin Plan objectives and other water quality investigations. After numerous years of testing, scientists have come to recognize that *C. dubia* is not always an ideal toxicity testing species, in particular when investigating the health of some of California's more saline inland surface waters. California has a number of water bodies, including the Sacramento-San Joaquin Delta, the Colorado River, the New River and the Alamo River, that frequently have conductivities which exceed the upper salinity tolerance of *C. dubia*. US EPA (2002) recommends a salinity of 1 ppt or a conductivity of approximately 1900 µS/cm, as the upper limit for use with freshwater test species. In-house tests have repeatedly demonstrated that *C. dubia* reproduction is negatively impacted by samples with conductivities above 2000 µS/cm (UCD ATL, unpublished data). For marine test species, on the other hand, USEPA (2002) recommends a lower salinity limit of 5 ppt. This creates a significant gap between the freshwater species' upper limit and the marine species' lower limit, resulting in a limited number of surrogate species appropriate for testing within this salinity range.

The amphipod species, *Hyalella azteca,* is tolerant of a broad conductivity range, which bridges the salinity gap between the US EPA freshwater and marine methods. A number of researchers have adapted the US EPA *H. azteca* 10-d sediment test for water column testing (Werner et al, 2008; SWRCB, 2002; Phillips, 2002). With an increasing interest in *H. azteca* as a water column test species, researchers need a refined testing protocol and a better understanding of the test's effectiveness in detecting water column toxicity. Three important factors contribute to one's ability to detect and identify a toxicant: the sensitivity of the species to toxicants, the method's ability to detect statistical differences when toxicants are present, and a researcher's ability to identify a toxicant using Toxicity Identification Evaluation (TIE) procedures. This paper compares the sensitivity of two water column toxicity tests, the chronic US EPA *C. dubia* survival and reproduction test, and a 10-d *H. azteca* survival and growth test.

The US EPA (2002) recommends selecting multiple species in different trophic levels for monitoring projects. Utilizing multiple species with varying chemical sensitivities will improve the likelihood that the presence of any toxicants will be detected. Among the species utilized in the US EPA freshwater manuals, *C. dubia* is more sensitive to pesticides than *Selenastrum capricornutum* or *Pimephales promelas*. Several authors have demonstrated that *H. azteca* is also relatively sensitive to pesticides (Werner et al, 2007; Ankley et al, 1995; Amweg et al, 2006; Burkepile et al, 2000). This sensitivity contributes to the increasing interest in developing standardized *H. azteca* chronic water column toxicity test methods. To evaluate these organisms' sensitivity to different chemicals, several tests were conducted with both *C. dubia* and *H. azteca* in two different water matrices in order to compare their effect concentration data. Pyrethroid pesticides were of particular interest in this study because their use is on the rise after several popular organophosphate pesticides were banned from use.

Percent minimum significant differences (PMSDs) give researchers an indication of the statistical sensitivity of test methods. PMSD is the smallest difference between the mean performance of a control and the mean performance of a sample can be detected by a given statistical test. The calculation of the PMSD differs depending on the statistical test being examined. This sensitivity is important in ambient monitoring projects as well as in the interpretation of TIEs, because the PMSD can indicate the reliability of a non-significant result. When a PMSD is small, most biologically significant differences in test organism performance will be detected by the statistical comparison, whereas in cases of a large PMSD, biologically significant differences in performance may not be detected by a statistical comparison. We compared PMSDs for both species to evaluate the relative statistical sensitivities of the two test methods.

With the increase in use of pyrethroid pesticides in urban and agricultural areas (Lee et al, 2002; Weston et al, 2006) and the recognition that *H. azteca* are relatively sensitive to this pesticide class, scientists are also interested in how well these chemicals can be tracked and fingerprinted throughout the TIE process. Previous studies have demonstrated that pyrethroid pesticides in particular have extremely high adsorption rates (Lee et al, 2002). While TIEs are generally initiated within 48 hours of sample toxicity, the amount of time hydrophobic compounds spend in storage can be a problem, as toxicant degradation and adsorption increase with time. A study was conducted to determine how much pesticide was lost to adsorption to sample container and test beaker walls. Understanding loss gives researchers some indication of what concentrations may be bioavailble to the test organism, especially under the extended storage time required for TIEs.

The US EPA TIE methods for *C. dubia* are well established (US EPA, 1992; US EPA, 1993). Generally these TIE procedures can be used for several species, but in many cases the individual manipulations have to be tested with unfamiliar species to understand the species' tolerance to various TIE chemicals. For example, repeated attempts at adding a range of concentrations of sodium thiosulfate to in-house *H. azteca* TIEs have often resulted in high mortality in method controls, even though this manipulation has been used successfully with *C. dubia*. Moreover, establishing a range of non-toxic chemical concentrations for TIEs can be even more difficult with varying values of hardness in ambient samples, as organism response differs as hardness increases.

Due to the high octanol-water partitioning coefficient, pyrethroids can be problematic when using columns that remove non-polar organic chemicals, such as C8 or C18 columns. Recovery in MeOH extraction can also be low, and as a result may not provide a strong enough signal in the TIE to interpret the results. Utilizing piperonyl buteoxide (PBO) as a TIE tool becomes important to add to the weight of evidence reflected in the TIE results. The PBO manipulation was implemented to reveal the presence of pesticides through the inhibitory effects of PBO on the toxicity of organophosphate insecticides and the synergistic effects of PBO on the toxicity of pyrethroid insecticides. There have been a number of PBO studies which have been used to evaluate its effect on *H. azteca* survival (Ankley et al, 1995; Amweg et al, 2006), but little has been assessed in relation to the growth endpoint. This paper includes two studies evaluating PBO as a TIE manipulation; one to determine the optimum PBO concentration to use in *H. azteca*

TIEs, and a second to determine how much pyrethroid toxicity is synergized by the addition of PBO. Growth data is presented whenever available.

During four years of extensive testing, *H. azteca* were exposed to ambient water samples from the Sacramento – San Joaquin Delta both with and without the addition of PBO. The most common differences in *H. azteca* performance seen with the addition of PBO were increases and decreases in weight. The presence of suspected pesticides was revealed by chemical analysis of a number of samples which had effects on the weight endpoint associated with the addition of PBO. In the present study, we examined the ability of the PBO manipulation to reliably reveal the effects of pesticides on *H. azteca* weight. The frequency of detection of pesticides by chemical analysis was compared between samples that showed a significant effect on *H. azteca* weight with PBO addition and those that showed no toxicological effect.

The objective of this paper is to determine whether the *H. azteca* 10-d water column test is a viable option for evaluating ecosystem health of those water bodies with conductivities above the tolerance level for EPA freshwater species, specifically *C. dubia*. This data contributes a small amount of information to the start of an extensive method validation process.

Materials and Methods

Toxicity Test Procedures

All tests were conducted at the University of California Aquatic Toxicology Laboratory (UCD-ATL), Davis, California, USA.

The *C. dubia* chronic toxicity test method strictly followed US EPA protocols (US EPA, 2002) with test duration ranging from 6 to 8 d depending on the time required to produce a third brood. *C. dubia* used in testing were obtained from in-house cultures. The control water for *C. dubia* used in effect concentration studies consisted of Sierra Spring or Drinking Water (DS Waters of America, USA) amended with dry salts to moderately hard specifications (US EPA, 2002). Test endpoints were survival and reproduction.

H. azteca were purchased from Aquatic Research Organisms (Hampton, NH, USA). Upon receipt, amphipods were moved to 10-L aquaria, fed, and acclimated to laboratory test conditions for 48 h. The 10-d testing procedure used in this monitoring study was based on protocols described in the Quality Assurance Management Plan for the State of California's Surface Water Ambient Monitoring Program (2002) and by US EPA (2000). At test initiation, water samples were shaken rigorously in original sampling containers, and sub-samples were filtered (ambient monitoring tests only) through a 53-μm screen to remove debris and other organisms. Water was then warmed to test temperature (23 ± 1^{o}C) in 600 ml beakers using a water bath maintained at 25±2^{o}C, and aerated at a rate of 100 bubbles/min until DO concentration was 4.9 - 8.9 mg/l. De-ionized water amended to US EPA moderately hard specifications (US EPA, 2000]) was used for controls. Tests were initiated with 9-14 d old *H. azteca*. Each of four replicate 250-ml glass beakers contained 100 ml of water, a small piece of nitex screen (approx. 4 cm^2) for use as substrate for the *H. azteca*, and 10 organisms. Animals were fed a mixture of yeast, organic alfalfa and trout chow (1 ml per replicate) at test initiation and on Days 2, 4, 5, 6, and 8. Tests

were conducted at 23 ± 1°C with a 16h:8h L:D photoperiod. Mortality was recorded daily and 80% of test water was renewed every other day. On day 10, the surviving *H. azteca* were dried to constant weight at 103-105 °C, and weighed using a Mettler AE 163 balance.

Analytical Chemistry

All analytical samples were collected in 1-L glass amber bottles and preserved with dichloromethane (DCM) within 24 hours of collection at the UCD ATL. Aqueous pesticide concentrations were determined by the California Department of Fish and Game Water Pollution Control Laboratory (Rancho Cordova, CA). Samples were extracted within 7 days of sample receipt. Water extractions followed USEPA Method 3510C – Separatory Funnel Liquid-Liquid Extraction. One-liter samples were fortified with the surrogates triphenyl phosphate and dibromooctaflurobiphenyl to monitor extraction efficiency and extracted twice with DCM using a mechanical rotating extractor. The extractions were dried using sodium sulfate, concentrated and solvent exchanged with petroleum ether using Kuderna-Danish evaporative glasswater equipped with a 3-ball snyder column followed with a micro-snyder apparatus and adjusted to a final volume of 2 ml in iso-octane. Final extracts were analyzed for OP pesticides using USEPA method 8141B, and for pyrethroids using USEPA method 8081B. OP pesticides were analyzed using a dual column high resolution gas chromatography with flame photometric detectors in phosphorous mode. Pyrethroids were analyzed using a dual column high resolution gas chromatography equipped with an electron capture detector.

Effect Concentration Tests

H. azteca test sensitivity to four current-use insecticides, the OPs, diazinon and chlorpyrifos, and the pyrethroids, bifenthrin and cyfluthrin was determined using standard effect concentrations (NOEC, LOEC, LC_{50} and EC_{25}). Analytical grade pesticides were obtained from ChemService (West Chester, PA). For each chemical, *C. dubia* and *H. azteca* were exposed to a minimum of five concentrations, a solvent control and a negative control in both a synthetic control and delta water series. The test waters consisted of the moderately hard synthetic waters described earlier or non-toxic 1.0 micron filtered Sacramento-San Joaquin Delta water (Byron, CA). Both dilution waters were amended to a specific conductance of 900 µS/cm with Instant Ocean® (Aquarium Systems, Inc., Mentor, OH) and a pH of 7.9 to better represent conductivities that are typically found in the Sacramento – San Joaquin Delta. Pesticide grade methanol was used as a carrier for the pesticides and did not exceed 0.05%. Samples of each pesticide concentration and the solvent control were collected at test initiation, immediately preserved with DCM, and submitted for analysis within 14 d. Lethal and sublethal effect concentrations were calculated using CETIS v.1.1.2 (Tidepool Scientific Software, McKinleyville, CA, 2006). NOEC and LOEC values were calculated using USEPA standard statistical protocols (USEPA, 2002). LC50 and EC25 concentrations were calculated using linear regression, non-linear regression or linear interpolation methods.

Sensitivity Comparison for Toxicity Endpoints

Percent minimum significant difference (PMSD) is the minimum difference in survival, fecundity or growth between an ambient sample and a control needed for statistical significance, expressed as the percentage of control performance for a given endpoint. PMSDs for lethal and sublethal endpoints were calculated for samples examined in ambient *H. azteca* 10-d water

column tests as part of the Pelagic Organism Decline project or *C. dubia* 7-d chronic tests as part of the Surface Water Ambient Monitoring Program (SWAMP) at UCD-ATL between January 2008 and June 2009. Samples were randomly selected among all toxic and non-toxic samples whose conductivity fell within the range optimal for the health of the test organisms. *C. dubia* data were exclusively from ambient water samples with a conductivity of ≤2,000 uS/cm. Two sets of data used for *H. azteca* tests were from samples with a conductivity <2,000 uS/cm, or <10,000 uS/cm. The first data set reflects the performance of the *H. azteca* test within the conductivity range suited for *C. dubia*, while the second data set reflects the wider range of conductivity over which *H. azteca* shows no detectable decrease in survival or weight (UCDATL, unpublished data).

Selected test data was compared to respective controls using US EPA standard single-concentration statistical protocols (US EPA, 2002). Fisher's exact test was used to detect differences in survival in the *C. dubia* chronic test. *H. azteca* survival was arcsine square root transformed before analysis. *C. dubia* reproduction, *H. azteca* survival, and *H. azteca* growth were examined by homoschedastic (same variance) t-tests, heteroschedastic (separate variance) t-tests or Wilcoxon tests depending on the normality of the distributions and homogeneity of variances of the comparison under consideration. Fisher's exact test minimum significant differences (MSDs) were calculated as the smallest difference in survival that would constitute a significant difference and thus indicate toxicity. MSDs of homoschedastic and heteroschedastic t-tests were determined following Wang et al. (2000) and Phillips et al. (2001). Wilcoxon test MSDs were calculated based on the method proposed by van der Hoeven (2008). A constant was added or subtracted from the performance of each replicate in the experimental treatment to change the ranks of the data such that the reduction in rank performance between the control and the experimental treatments was the minimum reduction detectable by the Wilcoxon test. All MSDs were converted to PMSDs by dividing the MSD by the mean control performance and multiplying the result by 100.

Toxicity Identification Evaluations

Three studies were conducted to address some of the emerging questions related to sample storage and the effectiveness of a specific TIE manipulation, the addition of PBO. The PBO manipulation has been used in TIEs for a number of years to evualuate the presence of pyrethroid and OP pesticides (Ankley et al, 1995; Amweg et al, 2006; Wheelock et al, 2006).

Insecticide Adsorption and Loss

To quantify the loss of chemical due to adsorption during storage and testing, a test was performed without organisms simulating typical procedures in a *H. azteca* test. Insecticides were spiked into laboratory control water (USEPA, 2000) and non-toxic, unfiltered ambient water from the Sacramento-San Joaquin Delta. Initial nominal concentrations were 50 ng/L for permethrin, bifenthrin, lambda-cyhalothrin, cyfluthrin and esfenvalerate; and 100 ng/L for chorpyrifos. These concentrations were selected because of their ecological relevance. The test solution was rigorously mixed, dispensed into 3.8-L low density polyethylene cubitainers, then stored in the dark at 4°C to mimic sample collection procedures. On the following day (test initiation), each cubitainer was shaken vigorously for 30 s just prior to pouring sample water into each of three 600 ml glass beakers, where water was aerated and warmed to the test temperature

of 23°C. Four replicate aliquots (100 ml) were subsequently poured from each 500 ml beaker into 250 ml glass beakers routinely used for toxicity testing, for a total of 12 replicates. Warming samples and pouring warmed samples into replicate beakers occurred on each day that a test renewal would typically occur.

Loss to Sample Container Storage: Samples for chemical analyses were collected immediately following pesticide spiking. These samples represented the initial concentration of pesticides, or what hypothetically might be collected from a water body at the time of sample collection. Additional samples, representing adsorption during storage, were also collected after 3, 4, 9 and 10 d of storage at 4°C. These samples also represent the concentrations that were poured into warming beakers on Days 2, 3, 8, and 9 of the test since the samples were stored for 24 hours before the simulated test was initiated.

Additional Loss to Toxicity Test Containers: To evaluate chemical fate during a toxicity test, samples for chemical analyses were collected from test containers (replicate beakers) on Day 4 and 10 of the simulated test, representing typical acute and chronic exposure durations for *H. azteca*. One-liter samples were collected by combining 80 mL of each of the 12 replicate beakers. All analytical samples were collected in 1 L glass amber bottles and preserved with 10 mL of DCM within 24 hours of sample collection.

Two test methods were evaluated, a daily renewal and an every other day renewal, in order to determine whether more frequent renewals would increase the bioavailability of pesticides to the test organism. Comparisons were also made between a synthetic moderately hard control water and an unfiltered ambient water using the every other day renewal method. This comparison was made to determine whether there were any matrices effects on adsorption.

Effectiveness of PBO as a TIE Tool

During an ambient monitoring project, 100 µg/L of PBO was initially used as a TIE manipulation because that concentration was well below the LC_{50} for *H. azteca* (UCDATL unreported data, Ankley et al, 1995) and it had been used successfully for many TIEs in *C. dubia* tests. It soon became apparent that this concentration negatively impacted *H. azteca* growth in the method blank. In-house tests revealed that 25 µg/L was generally a safe concentration for the growth endpoint, but whether this lower PBO concentration was effective as a TIE manipulation for higher pesticide concentrations, specifically with respect to the survival endpoint, was unknown. A test was initiated to evaluate how effective 25, 50 and 100 µg/L PBO was as a TIE manipulation for approximately one toxic unit[1] (TU) of three pesticides: permethrin, bifenthrin and chlorpyrifos. The TU concentration was determined from the nominal LC_{50} concentrations generated in the effect concentration tests. The test also included a solvent control with and without PBO. PBO was expected to accelerate the toxicity of the two pyrethroids and alleviate the toxicity of the OP pesticide. Following the test, each PBO treatment was statistically compared to its non-PBO counterpart using the Wilcoxon statistical test to determine whether PBO accelerated or decelerated toxicity on any given day during the test.

[1] A toxic unit is defined as the concentration of a specific chemical present in a sample divided by the 96-h LC_{50} concentration for the species of interest.

One additional *H. azteca* test was conducted to determine the potency of 25 µg/L PBO in synergizing toxicity with two pyrethroids pesticides, permethrin and bifenthrin. A minimum of four concentrations of each pesticide and a solvent control were tested with and without PBO. The pesticide concentrations ranged from 1/32 to 1 TU for each chemical. After test termination, effect concentrations were calculated, using CETIS software as described above, and compared to determine the extent to which toxicity was synergized by the presence of PBO.

Evaluation of the significance of the weight endpoint: In order to verify the ability of the PBO manipulation to reveal the effects of pesticides on *H. azteca* weight, a set of samples that caused a significant PBO effect and a set of samples that did not cause a PBO effect were analyzed for the presence of both pyrethroid and organophosphate insecticides. The results of the toxicological and chemical analyses were categorized as indicating the presence or absence of pesticide, and toxicological and chemical results were compared in a 2x2 contingency table by Fisher's Exact test. The null hypothesis was that no association existed between the results of toxicological and chemical analyses, and the alternate hypothesis was that chemical analysis indicated the presence of pesticides more often in the event of a significant PBO effect on *H. azteca* weight.

Results

Chemical Sensitivity

Table 1 lists the NOEC, LOEC, LC_{50} and available EC_{25} concentrations for both species exposed to each pesticide. The control of the *C. dubia* test conducted with cyfluthrin did not produce the minimum number of offspring (15 neonates/surviving female) that EPA requires to consider an acceptable test. This test only produced an average of 11.4 neonates/surviving female in the 900 µS/cm synthetic control water.

Statistical Sensitivity

H. azteca survival data were often non-normally distributed, and were consequently analyzed mostly using Wilcoxon tests. Whether tested by Wilcoxon test or t-test, *H. azteca* survival MSDs were low (Table 2), both for tests within the range of conductivity physiologically compatible with *C. dubia* (<2 mS/cm) and for the entire optimal range for the survival of *H. azteca* (<10 mS/cm).

For the sublethal endpoints of *H. azteca* and *C. dubia* the majority of test were analyzed using t-tests. PMSDs of *C. dubia* reproduction comparisons were low. PMSDs of *H. azteca* weight was highly variable, sometimes reaching above 100%.

Insecticide Adsorption and Loss During Storage and Testing

Table 3 lists the concentrations of six individual pesticides and the percent loss associated with adsorption to storage containers and test beakers.

Loss to Sample Containers: As an approximation, the pesticide losses from the six pesticides were averaged within a specific treatment to simplify quantification. In instances where calculating the average percent loss included a concentration below the detection limit, the lowest percent loss was used in the calculation. As such, these values may slightly under-

estimate the actual loss occurring under these conditions. The greatest loss occurred during the first 72 h of storage, regardless of the water type and losses generally increased with increasing storage time. One decrease in loss occurred between Day 8 and Day 9 in the control water series. Analytical variability may have played a role in this decrease, especially since concentrations were near the minimum detection limits for that analytical method. Adsorption was slightly less in the ambient water compared to the synthetic control after 3 and 9 days of storage with an average difference of 9.2 and 5.1 percent difference, respectively.

Additional Loss to Toxicity Test Containers: Analytical samples collected from the test replicate beakers represented the lowest pesticide concentration in a test at either the 96-h or 10-d time point. Concentrations were at their lowest, because the measurement included losses to multiple surfaces including the sample container, the warming beakers and the replicate beakers. The loss in all treatment types was approximately 93% in the replicate beakers by 96 hours in to the test. Losses for both water types were within 3.1% of each other at both the 96-h or 10-d test time point suggesting that any adsorption to particulate matter in the ambient water had little effect on bioavailability.

The average percent loss between the two water-renewal methods (daily versus every other day) in the control water series were within 2.1% of each other at both the 96-h and 10-d time points. This similarity suggests that organisms employed in toxicity tests are most likely exposed to similar chemical concentrations in either renewal method in relation to their original concentration.

In general, the maximum average pesticide concentrations on Day 2 of the test that organisms would see in a typical toxicity test are about one fifth of the concentrations that are originally present in a sample. This is most likely an overestimation, taking into consideration the likelihood that pesticides also bind to the warming beakers. By the end of the test, the minimum concentrations organisms are likely to encounter in a toxicity test are typically less than one twentieth of their original concentration.

Effectiveness of PBO as a TIE Tool

TIEs are more easily interpreted when the signal from any manipulation is more apparent. Daily organism survival in the test evaluating the efficacy of three concentrations of PBO is presented in Table 4. All concentrations of PBO accelerated toxicity of two pyrethroids and decelerated toxicity of the OP pesticide. No statistical differences occurred in the method blanks, suggesting that all three PBO concentrations were non-lethal and suitable for a TIE addressing the survival endpoint. Permethrin with all three concentrations of PBO were statistically different from the permethrin alone on Day 2 of the test only. The higher two concentrations of PBO, 50 and 100 µg/L, in the presence of bifenthrin, were also statistically different than bifenthrin alone on Days 2 – 4. In all cases, 25 µg/L PBO produced a less distinct TIE signal, either by duration or differences in % survival, than the 50 and 100 µg/L PBO. The weight endpoint with PBO could not be evaluated for permethrin and bifenthrin, because all organisms in treatments containing pyrethroids exhibited 100% mortality by the end of the test. With chlorpyrifos, the higher two concentrations of PBO reduced mortality for the final four days of the test, while the lowest concentration of PBO only showed statistical differences in the final two days of the test. Growth

in chlorpyrifos treatments, with and without PBO, was not statistically different, suggesting that PBO may not be an effective TIE tool at any concentration for the growth endpoint.

Table 5 summarizes results of *H. azteca* exposures to permethrin and bifenthrin in the presence of 25 µg/L PBO. The permethrin LC_{50} was potentiated by 2.48 and 8.81 fold for the 96-h and 10-d LC_{50}s, respectively. Synergism for bifenthrin was 3.95 and 3.22 fold for the 96-h and 10-d LC_{50}s, respectively. Little growth data was available to adequately address synergism in the growth endpoint.

Evaluation of the significance of the weight endpoint: The results of the Fisher's Exact Test conducted on ambient samples suggest that no association existed between the detection of either pyrethroid or OP pesticides and significant effects of 25 µg/L PBO on *H. azteca* growth. Pyrethroids were detected in 3 out of 11 samples that showed reduced weight with PBO addition, and they were detected in 4 out of 12 samples that did not show a significant PBO effect on weight (one-tailed Fisher's Exact Test, /P/ = 0.7776). OP pesticides were detected in 0 out of 10 samples that showed increased growth with PBO addition, and they were detected in 1 out of 12 samples that did not show a significant PBO effect on weight (one-tailed Fisher's Exact Test, /P/ = 1.000).

Discussion

Limited effect concentration data is available for *H. azteca* water column tests, rendering it difficult to make broad generalizations about which species is more sensitive to a particular class of pesticides. Furthermore, better comparisons are made between commonly used methods with standardized organism age, test conditions and test durations. Whenever possible, the publications referenced below were selected based on similarities in test duration and the use of an organism age that matches the life stages of standardized test methods.

H. azteca were far more sensitive to pyrethroid insecticides than *C. dubia* in our studies for both survival and sublethal endpoints. The LC_{50} concentrations for *H. azteca* (1.74 ng/L in synthetic control water and 1.86 ng/L in filtered delta water) for cyfluthrin were at least 382 times lower than those of *C. dubia* (703.8 and 712.3 ng/L). Yang and Hunter (2007) reported 96-h LC50 concentrations ranging from 0.093 to 0.210 µg/L to *C. dubia*. These concentrations are considerably lower than our 7-d *C. dubia* LC_{50} concentrations, but are considerably higher than our *H. azteca* LC_{50}s.

The bifenthrin LC_{50} concentrations for *H. azteca* (2.7 ng/L in synthetic control water and 2.3 ng/L in filtered delta water) were at least 115 times lower than those of *C. dubia* (344.9 and 266.1 ng/L). Liu (2005) reported a 96-h *C. dubia* LC_{50} for mixed isomers of cis-bifenthrin of 144 ng/L. Although this author reported varying sensitivities to different cis-bifenthrin isomers, the data supports the conclusion that *C. dubia* is less sensitive to bifenthrin than *H. azteca*.

Werner et al (2007) summarized the effects of pyrethroid insecticides on of numerous aquatic organisms and found that *H. azteca* was frequently more sensitive to pyrethroids by more than an order of magnitude than *C. dubia*. Van Wijingaarden et al. (2005) reported that amphipods were sensitive to repeated application of pyrethroids in microcosm and mesocosm studies in 100% of the studies where organisms were repeatedly exposed to 0.1 to 1 TU. In contrast, cladocerans

only were impacted in 50% of the studies. Both our data and published literature values strongly suggest that *H. azteca* is more sensitive to pyrethroids than *C. dubia*. Ambient monitoring programs targeting waterbodies that are likely to contain pyrethroid insecticides would benefit from the greater sensitivity of *H. azteca*, especially in samples with conductivities above 2 mS/cm.

In contrast, *C. dubia* were generally more sensitive to both OP insecticides, diazinon and chlorpyrifos, than *H. azteca* in our studies. The LC_{50} concentrations for *C. dubia* (167.5 ng/L in synthetic control water and 164.1 ng/L in filtered delta water) for diazinon were a minimum of 18 times lower than those of *H. azteca* (3080 and 4310 ng/L). The EC_{25} concentration for *C. dubia* for control water was approximately 8 times that of *H. azteca*. Effect concentrations determined here confirm those known from other studies. Giddings et al. (2000) reported the geometric mean 48-h *C. dubia* and *H. azteca* LC_{50} concentrations for diazinon of 0.49 and 22 µg/L, respectively. Burkepile et al. (2000) also reported 48-h *C. dubia* and *H. azteca* LC_{50} concentrations of 0.92 and 15.07 µg/L, respectively. Both the results of our study and the published literature values imply that *C. dubia* are more sensitive to diazinon than *H. azteca*.

The 7-d LC_{50} concentrations for *C. dubia* (23.7 ng/L in synthetic control water and 23.2 ng /L in filtered delta water) for chlorpyrifos were approximately one fourth of those for *H. azteca* (105.2 and 102.6 ng/L), suggesting that the cladoceran is the more sensitive species for the survival endpoint. Sublethal endpoints were more varied. The limited NOEC and LOEC data suggests that *H. azteca* was less slightly more sensitive than C. dubia in synthetic control water and less sensitive in filtered delta water. Ankley et al (1995) reported a 96-h LC_{50} concentration of 0.04 µg/L for *H. azteca*. For *C. dubia*, Bailey et al, (1996; 1997) reported a 96-h LC_{50} concentration of 0.05 and 0.06 µg/L, respectively. For chlorpyrifos, the published literature values suggest that *H. azteca* is more sensitive than *C. dubia* with respect to the survival endpoint, which contradicts our findings. These contradictions suggest that both species most likely have a similar sensitivity to chlorpyrifos.

Consideration of PMSDs is important in ambient monitoring programs because they directly affect test sensitivity. *H. azteca* survival data were analyzed using the Wilcoxon rank sum test in 28 out of 30 comparisons. This test is nonparametric, and MSDs are not usually calculated for nonparametric tests. The use of the method suggested by van der Hoeven (2008) for calculating the MSD of a Wilcoxon rank sum test made it possible to compare the statistical sensitivities of the *C. dubia* and *H. azteca* survival endpoints, and made it possible to include the results of Wilcoxon rank sum tests in the investigation of the statistical sensitivity of sub-lethal endpoints.

Weighted averages of mean PMSDs for each endpoint at conductivities below 2000 µS/cm delineate as follows: *H. azteca* survival (13.8%), *C. dubia* reproduction (17.2%), C. dubia survival (43.6%) and *H. azteca* growth (60.0%). PMSDs for *H. azteca* were nearly identical at conductivities between 2000 and 10,000 µS/cm. The PMSD for *C. dubia* 7-d survival (43.6%), one of the most commonly used endpoints in toxicity testing was significantly higher than that of H. azteca survival (13.8%). Even when the *C. dubia* chronic control showed 100% survival (10 out of 10 animals), the highest survival detectable in the experimental treatment as significantly different was 60% (PMSD = 40.0%). The *C. dubia* 7-d chronic test is unusual among commonly used aquatic toxicology tests in that every replicate includes only one animal. The survival data

generated by this test are therefore categorical in nature (each replicate was recorded as either alive or dead), and differences between treatments are evaluated using the Fisher's exact test, rather than the t-tests and the Wilcoxon test used to analyze the results of other endpoints. In contrast, replicates in the *H. azteca* 10-d water column test contain 10 individuals each, and survival data for each replicate is recorded as a percentage, making analysis by t-test or Wilcoxon test appropriate. *H. azteca* survival data were often non-normally distributed, and were therefore usually analyzed using Wilcoxon tests. The growth endpoint for *H. azteca* was far less sensitive (PMSD=60.0%) than the reproductive endpoint for *C. dubia* (PMSD=17.2%). The PMSD for *C. dubia* reproduction for the heteroschedastic t- test fell outside of the EPA's lower limit for an acceptable PMSD while the PMSDs for the homoschedastic t-test and Wilcoxon's test were within range, but very close to EPA's lower limit. The PMSD for the *H. azteca* growth endpoint may be higher due to the variable in organism size at the beginning of test initiation. To select organisms that are the right "age" for a toxicity test, commercial suppliers often pass *H. azteca* through specific mesh sized sieves in order to collect organisms that are 7 to 14 d old. In contrast, selection of *C. dubia* neonates is much more refined. Neonates are collected within an eight h window and used for testing before they are 24 h old in an effort to reduce between-replicate variability.

Our study demonstrated that pyrethroids as well as the OP chlorpyrifos in water samples present extraordinary challenges with respect to toxicity testing. During sample storage, the majority (approx. 80-88%) of insecticide was lost from solution within 72 h. These results corroborate data reported by Lee et al. (2002) who measured respective reductions of recovery of 37, 28, 21 and 32% of bifenthrin, *cis*-permethrin, *trans*-permethrin and deltamethrin in deionized water within the first 24 h of sample storage, with starting concentrations of 2 µg/L for each pyrethroid. Sharom and Solomon (1981) also reported adsorption of permethrin to borosilicate glass scintillation vials, with the majority of loss taking place within the first 24 h of contact. Contrarily, Hladik et al. (2009) reported only 5-7% loss of pyrethroids due to adsorption to cubitainers after storage for one week at 4 °C and one minute of shaking. The author spiked 400 ng/L of each pyrethroid. The thirty seconds of rigorous cubitainer shaking used in our study represents what is typically done during routine toxicity testing. Our results suggest that ambient samples must have pesticide concentrations substantially higher than any organisms' 96-h LC_{50} in order to elicit a toxic response within a 96 h testing period. In addition, the high rate of adsorption during sample storage suggests that extended storage times associated with TIEs typically result in less than 90% of the original pesticide concentrations remaining in the water sample. TIEs on samples strongly suspected to contain pyrethroids should therefore be limited to samples that are acutely toxic. We conclude that loss of non-polar chemicals due to adsorption may lead to gross under-estimation of toxicity experienced by organisms in the field when using laboratory toxicity tests.

The differences in adsorption, as average percent loss, between the daily and every-other-day renewal methods are 2.1 percent or less at both the 96-h and 10-d time points. This similarity suggests that either method may expose organisms to similar pesticide concentrations. Each storage time point also represents the pesticide concentrations just prior to pouring samples into the warming beakers of the simulated test. The difference in average percent loss in the storage containers between 72 and 96 h was a minimum of 4.7% suggesting that daily renewals may potentially provide the test organisms with concentrations slightly closer to the concentrations

that were present when a sample was originally collected. Unfortunately, data was not collected to evaluate the adsorption to the warming beakers prior to pouring a warmed aliquot into the replicate beakers. The highest concentrations in the replicate exposure beakers would be expected to occur immediately following renewal just prior to any adsorption to the replicate beaker walls. Wheelock et al. (2005) compared loss due to adsorption in acute *H. azteca* and *C. dubia* methods and found that even 4 h of holding permethrin prior to loading *C. dubia* resulted in a 50% loss of toxicity. The author reported the loss of toxicity to *H. azteca* was not as severe. Nonetheless, the pesticide loss to replicate container walls happened very quickly.

Similarly, the differences in percent loss in replicate beakers between the two water types at the 96-h and 10-d time points are 3.1% or less. These results suggest that *H. azteca* may be exposed to similar concentrations of pesticides, regardless of the water type. The loss to sample container walls in the ambient water was 9.2% less than in the control water, suggesting that organisms in an ambient sample test may be exposed to slightly higher pesticide concentrations initially, but these concentrations ultimately declined to match the concentrations in the control water series. These results coincide with those of Lee et al, (2002), whose overall reduction of recovery of pyrethroid pesticides in ambient water was higher than comparable deionized water samples. The largest factor that may increase the bioavailability of pyrethroids in ambient samples, especially in the first few days of the test, is adsorption of pesticides to suspended solids rather than the beaker walls. Pesticides may be even more bioavailable because the amphipod may cling to or consume these contaminated particles.

PBO proved to be an effective tool to synergize pyrethroid or antagonize OP toxicity for the survival endpoint. All three PBO concentrations tested increased or decreased survival as anticipated. Both 50 and 100 µg/L PBO accelerate the toxicity of pyrethroids and delayed the toxicity of OP pesticides without negatively affecting the positive control. More data is needed to determine whether PBO causes similar statistical differences in amphipod growth.

The magnitude of synergism or antagonism caused by the addition of PBO can affect researchers ability to interpret TIE results. Previous studies with *C. dubia* showed synergism up to 32 fold between 100 µg/L PBO and several pyrethroids (UCDATL, unpublished data). For *H. azteca*, the maximum increase in the toxicity of permethrin and bifenthrin with 25 µg/L PBO was 9 and 4 fold, respectively. Since the synergism in a *H. azteca* test is relatively low compared to the synergism in *C. dubia* studies, researchers may have a much smaller window of time within a TIE to observe an effect, making the TIE signal more difficult to interpret. For OPs administered at 1 TU, studies with *C. dubia* and other cladocera species have shown toxicity is delayed for at least 24 hours with the addition of 100 µg/L PBO (UCDATL, unpublished data, Ankley et al., 1991). In our study with *H. azteca*, the differences in survival following exposure to chlorpyrifos with or without PBO were more subtle and required statistical analyses of daily data to verify that PBO was effective.

Ankley et al. (1991) also demonstrated that higher concentrations of PBO were more effective at alleviating toxicity in higher concentrations of OP pesticides for three cladoceran species. Amweg et al. (2006) reported that higher concentrations of PBO synergized the toxicity of permethrin more than lower concentrations of PBO in *H. azteca* sediment tests. For both species, higher concentrations of PBO were more affective at eliciting the response one would expect in

from a TIE manipulation. Similar studies need to be done with *H. azteca* water column tests, especially to determine if TIE signals can be amplified to the point where they are readily apparent to the researcher. Stronger TIE signals ensure that researchers send analytical samples in for the most appropriate chemical class and expedite this decision making process.

No studies evaluating PBO efficacy as a TIE tool on the growth endpoint have been published to date. Our small study suggests that PBO at 25 µg/L is an ineffective TIE tool for the *H. azteca* growth endpoint. While this concentration does not affect growth in control treatments, 25 µg/L PBO does not adequately predict the presence of either insecticide class. Considering that LC_{50} concentrations are so close to the detection limits for pyrethroids, it is possible that concentrations affecting the growth endpoint are currently undetectable. A higher concentration such as 50 or 100 µg/L PBO can be used to ensure the TIE tool is effective for the survival endpoint.

Future Research

US EPA spent many years validating acute and chronic methods for *C. dubia* (US EPA, 2000). Since the *H. azteca* method described in this paper is relatively new, and additional authors have used dissimilar methods, validating any water column *H. azteca* method will require more studies to ascertain its reliability as a test method. Suggestions for future research include, but are not limited to:

- PBO, temperature (Weston et al, 2006; Ware and Whitacre, 2004), and esterase (Wheelock et al, 2006) have successfully been used as *H. azteca* TIE tools. More research is needed to address metal toxicity by addition of EDTA and sodium thiosulfate to provide a more complete set of TIE tools. To date, in-house studies have shown these manipulations can be problematic, especially when salinities are above 10 ppt.
- Acute *H. azteca* and *C. dubia* toxicity tests were utilized to evaluate five urban runoff samples from Roseville, CA (UCDATL, unpublished data). Although the source of toxicity was unknown, the *H. azteca* in four of the five samples died within 48 h while all *C. dubia* remained unaffected for the duration of the test. Similar studies should be performed to directly compare aggregate chemical sensitivity between both species. Research objectives should include investigations for various land use practices.

Conclusion

Based on the results of our study, we conclude that *H. azteca* is an appropriate surrogate species for toxicity testing in California, especially at water conductivities of 2-10 mS/cm. The 10-d *H. azteca* water column toxicity test has a number of advantages over the standardized US EPA chronic *C. dubia* test. These advantages include a much broader salinity tolerance of the test organism, greater sensitivity to a number of commonly used pyrethroids and better PMSDs on the survival endpoint. The higher sensitivity of *H. azteca* to some pesticides may enable researchers to initiate TIEs in a more timely fashion, thus minimizing problems associated with chemical loss during storage and testing.

The biggest disadvantage to using *H. azteca* is the lack of knowledge associated with TIE manipulations. Researchers should carefully weigh the financial risks associated with conducting

TIEs on samples that express toxicity late in the 10 day test period, especially since there is substantial pesticide loss due to adsorption and TIE tools are limited. Secondarily, the methods' growth endpoint is not a sensitive indication of sublethal toxicity due to significant data variability.

Overall, *H. azteca* is a good test organism where promulgated water column methods are not required, the conductivity range of samples falls between 200 and 10,000 µS/cm, and pyrethroids may be present in the sample. More studies are needed to further validate this method, however, it does have considerable promise based on its sensitivity.

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Table 1 a. Acute toxicity (measured) of organophosphate and pyrethroid insecticides to *C. dubia* and *H. azteca* in synthetic control water

Chemical	7-d Survival *C. dubia*				10-d Survival *H. azteca*			
	NOEC (ng/L)	LOEC (ng/L)	LC50 (ng/L)		NOEC (ng/L)	LOEC (ng/L)	LC50 (ng/L)	
			Estimate	95% C.I.			Estimate	95% C.I.
Diazinon	123	228	167.5	-	1140	2100	3080	2420 - 3670
Chlorpyrifos	20	28	23.7	-	66	128	105.2	88.2 - 141.7
Bifenthrin	288	432	344.9	323.4 – 352.7	2	3	2.7	2.5 - 3.0
Cyfluthrin	515	985	703.8	620 - 712	1.2	2.2	1.74	1.61 – 2.04

Table 1 b. Acute toxicity (measured) of organophosphate and pyrethroid insecticides to *C. dubia* and *H. azteca* in filtered delta water

Chemical	7-d Survival *C. dubia*				10-d Survival *H. azteca*			
	NOEC (ng/L)	LOEC (ng/L)	LC50 (ng/L)		NOEC (ng/L)	LOEC (ng/L)	LC50 (ng/L)	
			Estimate	95% C.I.			Estimate	95% C.I.
Diazinon	123	228	164.1	146.8 - 185.7	2800	5440	4310	4150 - 4460
Chlorpyrifos	20	28	23.2	22.4 - 23.7	66	133	102.6	91.2 - 114.7
Bifenthrin	179	288	266.1	233.2 - 323.4	1	3	2.3	1.6 - 4.5
Cyfluthrin	515	985	712.3	-	0.8	1.5	1.86	1.50 – 2.22

Table 1 c. Sublethal toxicity (measured) of organophosphate and pyrethroid insecticides to *C. dubia* and *H. azteca* in synthetic control water

Chemical	7-d Fecundity *C. dubia*				10-d Growth *H. azteca*			
	NOEC (ng/L)	LOEC (ng/L)	EC25 (ng/L)		NOEC (ng/L)	LOEC (ng/L)	EC25 (ng/L)	
			Estimate	95% C.I.			Estimate	95% C.I.
Diazinon	123	228	176.5	160.3 - 208	1140	2100	1410	<410 - 1990
Chlorpyrifos	20	28	21.9	21.2 - 22.1	14	>14	-	-
Bifenthrin	288	432	245.4	203 - 302	0.6	2	1.3	<0.6 - 2.3
Cyfluthrin*	268	515	351.6	240 - 412	2.2	3.7	-	-

* reproductive data for C. dubia was excluded because the average reproduction in the control did not meet EPA's minimum reproductive requirement for test acceptability.

Table 1 d. Sublethal toxicity (measured) of organophosphate and pyrethroid insecticides to *C. dubia* and *H. azteca* in filtered delta water.

Chemical	7-d Fecundity *C. dubia*				10-d Growth *H. azteca*			
	NOEC (ng/L)	LOEC (ng/L)	EC25 (ng/L)		NOEC (ng/L)	LOEC (ng/L)	EC25 (ng/L)	
			Estimate	95% C.I.			Estimate	95% C.I.
Diazinon	228	560	176.2	20.5 - 264.9	1140	2800	>2800	-
Chlorpyrifos	20	28	21.3	9.13 - 22.1	66	>66	-	-
Bifenthrin	179	288	231.9	211.6 - 262.9	<1	1	0.5	0.4 – 0.7
Cyfluthrin	<108	108	273.7	206 - 321	3.1	>3.1	-	-

a: Low weight of solvent control indicates that EC25 estimate may be an artifact of high weights in low concentration treatments.

Table 2. Percent minimum significant differences (PMSD) in *C. dubia* 7-d and *H. azteca* 10-d toxicity tests.

Species	Endpoint	Statistical Test Method	*N*	PMSD (%)		
				Mean	SD	Range
C. dubia	Survival	Fishers Exact	32	43.6	7.1	40 - 62.5
	Reproduction	Heteroschedastic T-test	3	12.2	3.1	9.9 - 15.7
		Homoschedastic T-test	23	17.7	10.7	5.8 - 41.1
		Wilcoxon	6	18.0	7.0	7.0 - 25.3
H. azteca (<10 mS/cm)	Arcsinsqrt Survival	Homoschedastic T-test	2	13.1	0.0	13.1 - 13.1
		Wilcoxon	28	13.1	8.4	3.5 - 32.4
	Weight	Homoschedastic T-test	35	55.8	33.1	11.4 - 188.6
		Wilcoxon	3	61.6	17.7	48.1 - 81.6
H. azteca (<2 mS/cm)	Arcsinsqrt Survival	Homoschedastic T-test	2	13.1	0.0	13.1 - 13.1
		Wilcoxon	25	13.9	8.6	3.5 - 32.4
	Weight	Homoshedastic T-test	27	59.8	35.2	11.4 - 188.6
		Wilcoxon	3	61.6	17.7	48.1 - 81.6

Table 3. Loss of insecticide during storage and testing in 10-d *H. azteca* water column toxicity tests with different water renewal regimes. Nominal initial concentrations were 50 ng/L for pyrethroids and 100 ng/L for chlorpyrifos.

Chemical	Measured Initial Concentration (ng/L) [a]	Loss of Chemical as % of Initial Concentration After					
		3 d [b]	4 d [b]	5 d [c] (Day 4 of Test)	9 d [b]	10 d [b]	11 d [c] (Day 10 of Test)
Daily Water Renewal							
Chlorpyrifos	68	82.4	>95.6	>95.6	>95.6	>95.6	>95.6
Bifenthrin	21	80.8	80.8	88.5	92.3	92.3	96.2
Cyfluthrin	33	87.9	90.9	93.9	97.0	93.9	98.2*
Esfenvalerate	36	86.1	88.9	88.9	94.4	94.4	97.2
Lambda Cyhalothrin	36	86.1	88.9	88.9	91.7	88.9	94.4
Permethrin, Cis and Trans	54	81.5	87.0	90.7	>95.2	96.5*	94.4
Every Other Day Renewals							
Chlorpyrifos	68	82.4		>95.6	>95.6		>95.6
Bifenthrin	21	80.8		92.3	92.3		92.3
Cyfluthrin	33	87.9		93.9	97.0		93.9
Esfenvalerate	36	86.1		91.7	94.4		94.4
Lambda Cyhalothrin	36	86.1		94.4	91.7		94.4
Permethrin, Cis and Trans	54	81.5		92.6	>95.2		96.3
Delta Water: Every Other Day Renewals							
Chlorpyrifos	72	80.6*		>95.8	>95.8		>95.8
Bifenthrin	21	61.9		>99.0	90.5		>99.0
Cyfluthrin	28	78.6		>98.6	92.9		>98.6
Esfenvalerate	28	71.4		85.7	85.7		>99.3
Lambda Cyhalothrin	30	76.7		90.0	96.7		99.0*
Permethrin, Cis and Trans	36	77.8		95.6	78.9		>96.7

* concentration was below reporting limit
> concentration was below minimum detection limit
[a] chemical concentration immediately after spiking
[b] percent loss after storage in cubitainer and prior to pouring an aliquot into 600 mL warming beakers
[c] percent loss associated after storage and 1-2 d testing

Table 4. Concentration dependent effect of PBO on acute toxicity of nominal concentrations of permethrin, bifenthrin and chlorpyrifos to *H. azteca*.

Treatment	Mean Survival (%)									
	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Day 8	Day 9	Day 10
Control	100	100	100	100	100	100	100	100	100	100
Control + MeOH @ 0.02%	100	100	100	100	100	100	100	100	100	100
Control + MeOH + 50 µg/L PBO	100	100	100	100	100	100	100	100	100	98
Control + MeOH + 100 µg/L PBO	100	100	98	98	98	95	95	95	95	93
94 ng/L Permethrin	90	58+	23+	23+	18+	15+	8+	8+	5+	5+
94 ng/L Permethrin + 25 µg/L PBO	90	20*	0	0	0	0	0	0	0	0
94 ng/L Permethrin + 50 µg/L PBO	83	0*	0	0	0	0	0	0	0	0
94 ng/L Permethrin +100 µg/L PBO	73	0*	0	0	0	0	0	0	0	0
10 ng/L Bifenthrin	100	100	48+	20+	13+	8+	3+	0+	0+	0+
10 ng/L Bifenthrin + 25 µg/L PBO	100	98	0*	0*	0	0	0	0	0	0
10 ng/L Bifenthrin + 50 µg/L PBO	100	48*	5*	0*	0	0	0	0	0	0
10 ng/L Bifenthrin + 100 µg/L PBO	95	43*	0*	0*	0	0	0	0	0	0
143 ng/L Chlorpyrifos	100	100	100	100	100	69+	28+	23+	13+	10+
143 ng/L Chlorpyrifos + 25 µg/L PBO	100	100	100	100	98	87	71*	71*	50*	45*
143 ng/L Chlorpyrifos + 50 µg/L PBO	100	98	98	98	95	87	79*	79*	79*	74*
143 ng/L Chlorpyrifos + 100 µg/L PBO	100	100	100	100	100	97*	92*	92*	87*	87*

*: $P < 0.05$ with corresponding non-PBO chemical spike

+: $P < 0.05$ with Control + MeOH

Table 5. Synergistic effect of PBO on 96-h and 10-d *H. azteca* toxicity endpoints for permethrin and bifenthrin.

Toxicity Endpoint	Chemical	NOEC (ng/L)	Fold Increase Due to PBO	LOEC (ng/L)	Fold Increase Due to PBO	LC50/EC25 (ng/L)	Fold Increase Due to PBO
96-h Survival	Permethrin	47	2x	94.0	2x	76.9	2.5x
	Permethrin + 25 µg/L PBO	23.5		47		31.0	
10-d Survival	Permethrin	23.5	8x	47.0	8x	52.9	8.8x
	Permethrin + 25 µg/L PBO	2.94		5.88		6.0	
10-d Growth	Permethrin	47	8x	>47	n/a	14.9[a]	n/a
	Permethrin + 25 µg/L PBO	5.88		>5.88		-	
96-h Survival	Bifenthrin	5	4x	10	4x	15.8	4.0x
	Bifenthrin + 25 µg/L PBO	1.25		2.5		4.0	
10-d Survival	Bifenthrin	1.25	2x	2.5	2x	2.9	3.2x
	Bifenthrin + 25 µg/L PBO	0.625		1.25		0.9	
10-d Growth	Bifenthrin	2.5	2x	>2.5	n/a	-	n/a
	Bifenthrin + 25 µg/L PBO	1.25		>1.25		-	

a Unreliable estimate because dose-response was not consistent with a single trendline and weights of some treatments were anomalous.

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Linking mechanistic and behavioral responses to sublethal esfenvalerate exposure in the endangered delta smelt; *Hypomesus transpacificus* (Fam. Osmeridae)

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Abstract

Background: The delta smelt (*Hypomesus transpacificus*) is a pelagic fish species listed as endangered under both the USA Federal and Californian State Endangered Species Acts and considered an indicator of ecosystem health in its habitat range, which is limited to the Sacramento-San Joaquin estuary in California, USA. Anthropogenic contaminants are one of multiple stressors affecting this system, and among them, current-use insecticides are of major concern. Interrogative tools are required to successfully monitor effects of contaminants on the delta smelt, and to research potential causes of population decline in this species. We have created a microarray to investigate genome-wide effects of potentially causative stressors, and applied this tool to assess effects of the pyrethroid insecticide esfenvalerate on larval delta smelt. Selected genes were further investigated as molecular biomarkers using quantitative PCR analyses.

Results: Exposure to esfenvalerate affected swimming behavior of larval delta smelt at concentrations as low as 0.0625 $\mu g.L^{-1}$, and significant differences in expression were measured in genes involved in neuromuscular activity. Alterations in the expression of genes associated with immune responses, along with apoptosis, redox, osmotic stress, detoxification, and growth and development appear to have been invoked by esfenvalerate exposure. Swimming impairment correlated significantly with expression of aspartoacylase (ASPA), an enzyme involved in brain cell function and associated with numerous human diseases. Selected genes were investigated for their use as molecular biomarkers, and strong links were determined between measured downregulation in ASPA and observed behavioral responses in fish exposed to environmentally relevant pyrethroid concentrations.

Conclusions: The results of this study show that microarray technology is a useful approach in screening for, and generation of molecular biomarkers in endangered, non-model organisms, identifying specific genes that can be directly linked with sublethal toxicological endpoints; such as changes in expression levels of neuromuscular genes resulting in measurable swimming impairments. The developed microarrays were successfully applied on larval fish exposed to esfenvalerate, a known contaminant of the Sacramento-San Joaquin estuary, and has permitted the identification of specific biomarkers which could provide insight into the factors contributing to delta smelt population decline.

Background

The delta smelt (*Hypomesus transpacificus*) is a pelagic fish species endemic to the Sacramento-San Joaquin estuary, whose abundance has dramatically declined since the 1980s, and more precipitously in recent years [1-4]. It was listed as endangered in 2009, under both the Federal Endangered Species Act (ESA) and California Endangered Species Act (CESA). Considerable efforts are presently being made to understand the causes of this recent population decline [4,5], especially because several other pelagic species have shown similar population trends. Delta habitats have been compromised by a number of complex factors, both known and unknown, potentially affecting aquatic species throughout the Sacramento-San Joaquin watersheds and estuary [4]. Pollution, in the form of chemicals contained in runoff from agricultural and urban areas, and old mining sites, treated wastewater effluent, along with the effects of water exports, invasive species and habitat destruction are amongst potential causes for the population decline of several pelagic species [5].

Identifying the sublethal impacts of environmental stressors and their mechanistic effects on resident individuals and populations is a major challenge in ecotoxicology. Contaminants may not only affect organism survival, but can compromise ecological fitness of individual species via sublethal physiological, behavioral or immunological effects (e.g. [6-10]), consequently altering food web and ecosystem dynamics. However, such physiological endpoints are often difficult to determine in field studies, because they either require behavioral observation and measurements, or because affected individuals will not survive in the wild. Similarly, widely used ecotoxicological tools such as standard toxicity tests [11,12] cannot easily be adapted to resident species of concern, and, conversely, it is problematic to extrapolate test results obtained with surrogate species to resident species of concern [13]. Recent comparative studies have demonstrated a need for identifying effects directly in the species of concern, as traditional model organisms may differ in sensitivity and physiological response to environmental contaminants and other stressors [14,15].

Carefully selected molecular biomarkers can provide species-specific and sensitive, mechanistic information on the overall health of an organism, as toxic responses are often preceded by alterations in gene expression [16,17]. In particular microarray gene profiling is a powerful tool for defining genome-wide effects of environmental change on biological function [16,18,19]. The predictive value of microarrays as screening tools, as well as our understanding of these responses and their application in the field of ecotoxicology is rapidly growing. This technology can be applied in vertebrates and invertebrates, plants, algae, cell lines and unicellular organisms [20]. In addition, links are being established between specific molecular biomarkers identified by microarray technology, and higher-level life history parameters, such as metabolism, growth and reproduction [16,18,21,22]. Gene expression studies carried out over short-term exposures have allowed for the prediction of chronic stressor effects, such as reduced fecundity and embryonic arrest, somatic growth, and population dynamics [16,18,21,23]. Thus, specific gene responses in studied organisms would not only be indicative of health status, but when used in conservation studies, could highlight potential causes for population decline. However, few biomarkers are currently understood well enough to provide conclusive evidence of contaminant impacts on aquatic species in field monitoring, and extrapolating effects seen at the biomarker level to individual or population-level toxicity continues to be a challenge. For molecular biomarkers to be used as successful monitoring tools of individual, population and ecosystem damage, strong links need to continue to be established between gene expression and health status.

To better understand the sublethal effects of contaminants upon *H. transpacificus*, and to identify biomarkers for future field investigations, we have constructed a microarray with 8,448 Expressed Sequence Tags (ESTs). No genomic information was available on any database at the time this project began, other than a few mitochondrial sequences used in taxonomic studies [24]. We describe here, the construction and first application of this tool to identify genes in the delta smelt, specifically responding to exposure to esfenvalerate, a pyrethroid insecticide, and

present gene expression quantitation of selected biomarkers, utilizing these to explain observed swimming abnormalities. We used esfenvalerate in our study because biochemical responses and adverse effects on the whole organisms are relatively well understood [25] and therefore would aid interpretation of results in this "proof of principle" test. Esfenvalerate [(*S*)-a-cyano-3-phenoxybenzyl-(*S*)-2-(-4-chlorophenyl)-3-methylbutyrate] is a synthetic pyrethroid insecticide, widely used in agriculture, with a high risk to aquatic organisms [26]. It causes neurological damage by blocking sodium and potassium channels, resulting in repetitive neurological discharge [25]. In addition, pyrethroid insecticides are highly soluble in myelin sheaths of nerves, causing demyelination, resulting in conduction deficiencies through nerve lesions [27], directly affecting swimming ability, and impinging on foraging and migration. Fish are highly sensitive to this insecticide, with for example effects on bluegill behavior at measured concentrations as low as 0.025 $\mu g/L^{-1}$[28]. Pyrethroids have also been reported to affect growth, induce immune responses, reduce hepatic glycogen levels and delay spawning [9,29].

The main focus of this study was not the development of the microarray, rather the identification of molecular biomarkers specific to the delta smelt and stressors found in the San Joaquin-Sacramento delta. We present here results from annotated genes identified through microarray analyses and specifically quantitative PCR analyses of selected molecular biomarkers.

Results and Discussion

Effects of esfenvalerate exposure: mortality and swimming behavior

Fish larvae are known to be highly sensitive to esfenvalerate, with effects on swimming performance and enhanced susceptibility to predation resulting from concentrations as low as 0.0625 $\mu g/L^{-1}$[10]. Behavior alterations are construed as being consequential to the reported neurological mode of action of this pesticide, further affecting foraging, migration and reproduction [30]. Toxicity of pyrethroids in the Sacramento-San Joaquin estuary is likely alleviated by the presence of particles and organic matter, and to date concentrations of esfenvalerate detected in the water column were low, however, concentrations in winter storm runoff from agricultural lands have been reported up to 0.093 $\mu g.L^{-1}$ [31], influencing our decision to investigate dose response exposures to both high and environmentally relevant concentrations in confirmatory studies.

In terms of mortality, 10-d old delta smelt were only slightly more sensitive in this study ($LC_{50,24\ h}$ = 0.19 $\mu g.L^{-1}$) than 52-d old ($LC_{50,24\ h}$ = 0.24 $\mu g.L^{-1}$), however swimming performance of the younger larvae was affected at a concentration approximating one third of that observed affecting older fish (figure 1). Swimming abnormality in 10-d old larvae, intensified with increasing esfenvalerate concentration at 4 h, escalating significantly after 24 h exposure (figure 1a). This swimming abnormality was also concentration dependent in 52-d old fish, however swimming effects resulting from different time point measurements differed only at the highest exposure concentration of 0.250 $\mu g.L^{-1}$ (figure 1b), where effects on motion increased from 22.5% anomaly at 4 h to 45% at 24 h. Behavioral abnormalities, reduced food intake and growth, as well as increased susceptibility to predation were reported in fathead minnow larvae exposed to esfenvalerate for 4 h to concentrations above 0.455 $\mu g.L^{-1}$ [10]. Significant swimming impairments were determined in this study at 0.250 $\mu g.L^{-1}$, thus delta smelt are highly sensitive to sublethal esfenvalerate exposure. Furthermore, bioaccumulation in rainbow trout have resulted in concentrations 400 times higher than background ambient levels http://extoxnet.orst.edu.

Microarray application and q-PCR

Through the application of the delta smelt microarrays, and combined analytical methods, we have identified 288 ESTs, from which a number of genes of interest could be used to measure the effect of esfenvalerate and potentially other pyrethroid insecticides; further investigating their use as biomarkers in this species. Of the sequenced ESTs that responded significantly, 118 genes were successfully annotated; 170 matched unnamed or hypothetical proteins, or did not match the described annotation selection criteria; i.e. BLASTx searches resulted in expect-values greater than 1×10^{-5} and scores below 50. Based on gene ontology; molecular function, biological processes and cellular components, 94 unique genes were functionally classified (figure 2 and table 1) and described below. Based on the proportions of ESTs responding to successfully annotated unique genes (33%), it is estimated that of the 8,448 ESTs printed on the microarray, there could be above 2,500 unique genes identified in the delta smelt. These numbers however represent responses to a single contaminant, and should not be construed as final as there will have an intrinsic bias exerted upon them, however, the proportion of repeated sequences in the analyses was very low, with a maximum of nine repetitions for CHK1 checkpoint homologue and not more than one or two duplication for a few others. It is also important to note that the microarray was manufactured with incomplete genome data, thus information presented in figure 2 represents proportions of a limited number of available genes.

Differences between methods used allow for greater mining of possible biomarkers. The method by Loguinov *et al* [32] identified a single differentially expressed gene at 0.125 $\mu g/L^{-1}$ esfenvalerate (with no significant homol-

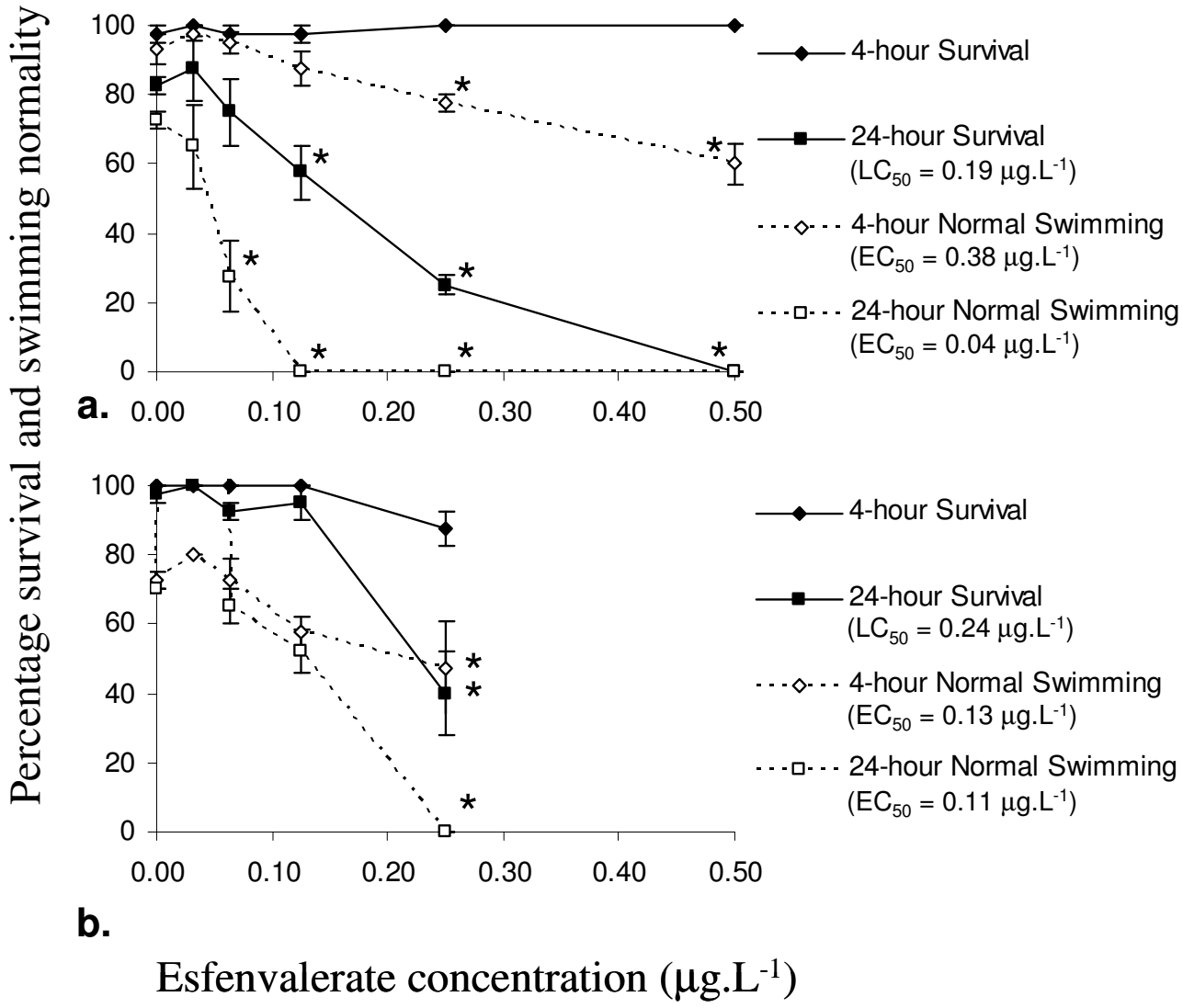

Figure 1
Swimming behavior and mortality. Percentage swimming normality and survival in (a.) 10-d old and (b.) 52-d old *H. transpacificus* exposed to esfenvalerate, ± standard errors (n = 10). * Indicates significant reduction in survival or swimming performance compared to solvent control.

ogy). The addition of LIMMA GUI analysis, also identifying this single gene, generated a broader list of genes for biomarker selection.

A large percentage of uniquely annotated genes, responding to esfenvalerate exposure; 49%, were classified as involved in various biological processes. These included genes encoding for ribosomal proteins, t-RNA synthases, telomerases, uncoupling proteins and genes involved in chromosome maintenance. Of greater interest was the identification of genes involved in neuromuscular activity; representing 19% of identified sequences, a further 12% eliciting immune responses, along with 6% related to oxidative stress, respiration and iron storage and 5% relevant to apoptosis. Digestion appears to have also been affected, along with growth and development, repre-

Table 1: Classification of differential gene expression of esfenvalerate responding genes in 52-d old *H. transpacificus* following 24 h exposure as identified by microarray analyses.

BLASTX top hit	Species match	Accession No.	Score	E-value	Concentration	Fold Change (+/-)	P-VALUE (or cut-off)	
Neuromuscular								
titin a	*Danio rerio*	ABG48500	110	4.00E-23	0.0625	1.3277	0.0625	**
smooth muscle cell-specific protein SM22 alpha	*Epinephelus coioides*	ABW04145	349	1.00E-94	0.1250	1.2252	0.0029	
toxin-1	*Oncorhynchus mykiss*	AAM21198	116	5.00E-25	0.1250	1.1884	0.0044	
thymosin beta-12	*Lateolabrax japonicus*	P33248	80	2.00E-13	0.1250	1.1876	0.0003	
similar to 19.9 kD myosin light chain isoform 1	*Danio rerio*	XP_685183	332	1.00E-89	0.1250	1.1720	0.0046	
ictacalcin	*Ictalurus punctatus*	AAY86967	145	1.00E-33	0.1250	1.1607	0.0021	
tropomyosin	*Theragra chalcogramma*	BAC44994	281	2.00E-74	0.1250	1.1368	0.0090	
N-acylsphingosine amidohydrolase	*Takifugu rubripes*	AAM43813	367	e-100	0.1250	1.1196	0.0082	
alanine-glyoxylate aminotransferase	*Platichthys flesus*	CAH59400	345	2.00E-93	0.1250	1.0860	0.0059	
titin b	*Danio rerio*	ABG48499	65	5.00E-09	0.1250	-1.0841	0.0045	
alpha-2,8-polysialyltransferase IV	*Oncorhynchus mykiss*	BAC77411	70	1.00E-10	0.1250	-1.0878	0.0030	
hedgehog acyltransferase-like, a	*Danio rerio*	NP_957181	295	1.00E-78	0.1250	-1.1419	0.0074	
parvalbumin	*Cyprinus carpio*	CAC83659	173	7.00E-42	0.1250	-1.1675	0.0033	
BTEB transcription factor	*Pimephales promelas*	ABO28528	107	1.00E-21	0.0625	-1.2019	0.0625	*
myosin regulatory light chain 2	*Salmo salar*	CAD89610	330	7.00E-89	0.1250	-1.2121	0.0020	
similar to Clca1 protein	*Danio rerio*	XP_694323	198	2.00E-49	0.0625	-1.2547	0.0625	*
ependymin	*Perca flavescens*	ABU49423	168	2.00E-40	0.0625	-1.2602	0.0625	*
aspartoacylase	*Danio rerio*	NP_001103573	384	e-105	0.0625	-1.3905	0.0625	*
Immune								
carboxypeptidase B	*Paralichthys olivaceus*	BAC53789	365	2.00E-99	0.1250	1.2897	0.0041	
fish-egg lectin (FEL)	*Cyprinus carpio*	P68512	192	2.00E-47	0.1250	1.1769	0.0003	
procathepsin B	*Oncorhynchus mykiss*	AAK69705	346	1.00E-93	0.0625	-1.1727	0.0625	*
gamma-glutamyl hydrolase	*Danio rerio*	NP_998487	223	6.00E-57	0.0625	-1.1939	0.0625	*
membrane glycoprotein	*Human coronavirus*	ABD75532	53	1.00E-05	0.0625	-1.1945	0.0625	*
beta-2 microglobulin	*Salmo salar*	AAG17525	176	8.00E-43	0.1250	-1.1995	0.0034	
microtubule-associated protein 1 light chain 3 alpha	*Danio rerio*	NP_999904	238	3.00E-61	0.0625	-1.2245	0.0625	*
microtubule-associated protein, RP/EB family, member	*Danio rerio*	NP_998805	272	1.00E-71	0.0625	-1.2298	0.0625	*
T-cell receptor beta chain ANA 11, putative	*Brugia malayi*	EDP38115	63	2.00E-08	0.1250	-1.2600	0.0052	
glycerophosphodiester phosphodiesterase domain containing 1	*Danio rerio*	NP_001004118	322	2E-86	0.0625	-1.2652	0.0625	*
CHK1 checkpoint homolog	*Xenopus tropicalis*	CAJ83813	92	2.00E-17	0.1250	-1.3981	0.0013	
Apoptosis								
tissue inhibitor of metalloproteinase 2	*Oncorhynchus mykiss*	AAU14867	265	3.00E-69	0.1250	1.1235	0.0082	
cathepsin H	*Danio rerio*	NP_997853	300	5.00E-80	0.1250	1.0898	0.0069	
caspase-3	*Dicentrarchus labrax*	ABC70996	223	6.00E-68	0.1250	-1.0647	0.0022	

Table 1: Classification of differential gene expression of esfenvalerate responding genes in 52-d old *H. transpacificus* following 24 h exposure as identified by microarray analyses. *(Continued)*

caspase-1	*Dicentrarchus labrax*	ABB05054	79	3.00E-13	0.1250	-1.1270	0.0061	
cathepsin S-like	*Oncorhynchus mykiss*	AAV32964	291	1.00E-77	0.1250	-1.1507	0.0047	
Redox and metal ion binding								
hydroxymethylbilane synthase	*Danio rerio*	CAM15096	369	e-101	0.1250	1.3333	0.0042	
hemopexin	*Danio rerio*	NP_001104617	313	2.00E-83	0.1250	1.2968	0.0052	
transferrin	*Salvelinus fontinalis*	BAA84100	326	1.00E-87	0.1250	1.1769	0.0080	
similar to leprecan-like 1 protein	*Danio rerio*	XP_695073	183	1.00E-44	0.1250	1.1543	0.0036	
similar to synaptic glycoprotein SC2	*Danio rerio*	XP_693420	430	e-119	0.1250	-1.1340	0.0073	
similar to LOC407663 protein	*Danio rerio*	XP_698537	124	6.00E-27	0.0625	-1.2429	0.0625	*
Growth and development								
yghl1 (Putative growth hormone like protein-1)	*Seriola quinqueradiata*	BAB62526	153	1.00E-35	0.1250	-1.1388	0.0072	
ZPA domain containing protein	*Oryzias latipes*	NP_001098216	168	3.00E-40	0.0625	-1.1568	0.0625	*
Detoxification								
pregnane × receptor	*Oncorhynchus mykiss*	ABP38412	206	2.00E-51	0.0625	-1.2321	0.0625	*
Osmotic stress								
hyperosmotic glycine rich protein	*Salmo salar*	AAO32675	134	8.00E-30	0.1250	-1.1053	0.0021	
Digestion								
similar to Apoa4 protein isoform 2 (Apolipoprotein)	*Danio rerio*	XP_698920	296	7.00E-79	0.1250	1.4959	0.0056	
chymotrypsinogen 2-like protein	*Sparus aurata*	AAT45254	460	e-128	0.1250	1.4211	0.0095	
pancreatic carboxypeptidase A1 precursor copy 2	*Tetraodon nigroviridis*	AAR16321	242	9.00E-63	0.1250	1.3204	0.0025	
pancreatic protein with two somatomedin B domains	*Paralichthys olivaceus*	BAA88246	214	e-100	0.1250	1.1297	0.0021	
chitinase (Zgc:55941)	*Danio rerio*	AAH44549	369	e-100	0.1250	-1.0618	0.0097	

Genes selected for quantitative PCR analyses are shown in bold. * indicates genes responding at 0.0625 μg.L^{-1} esfenvalerate and ** indicates gene responding to both 0.0625 and 0.125 μg.L^{1} esfenvalerate. Remaining genes responded only at 0.125 μg.L^{1} esfenvalerate exposure. Information represents proportions of a limited number of available genes on the applied microarray.

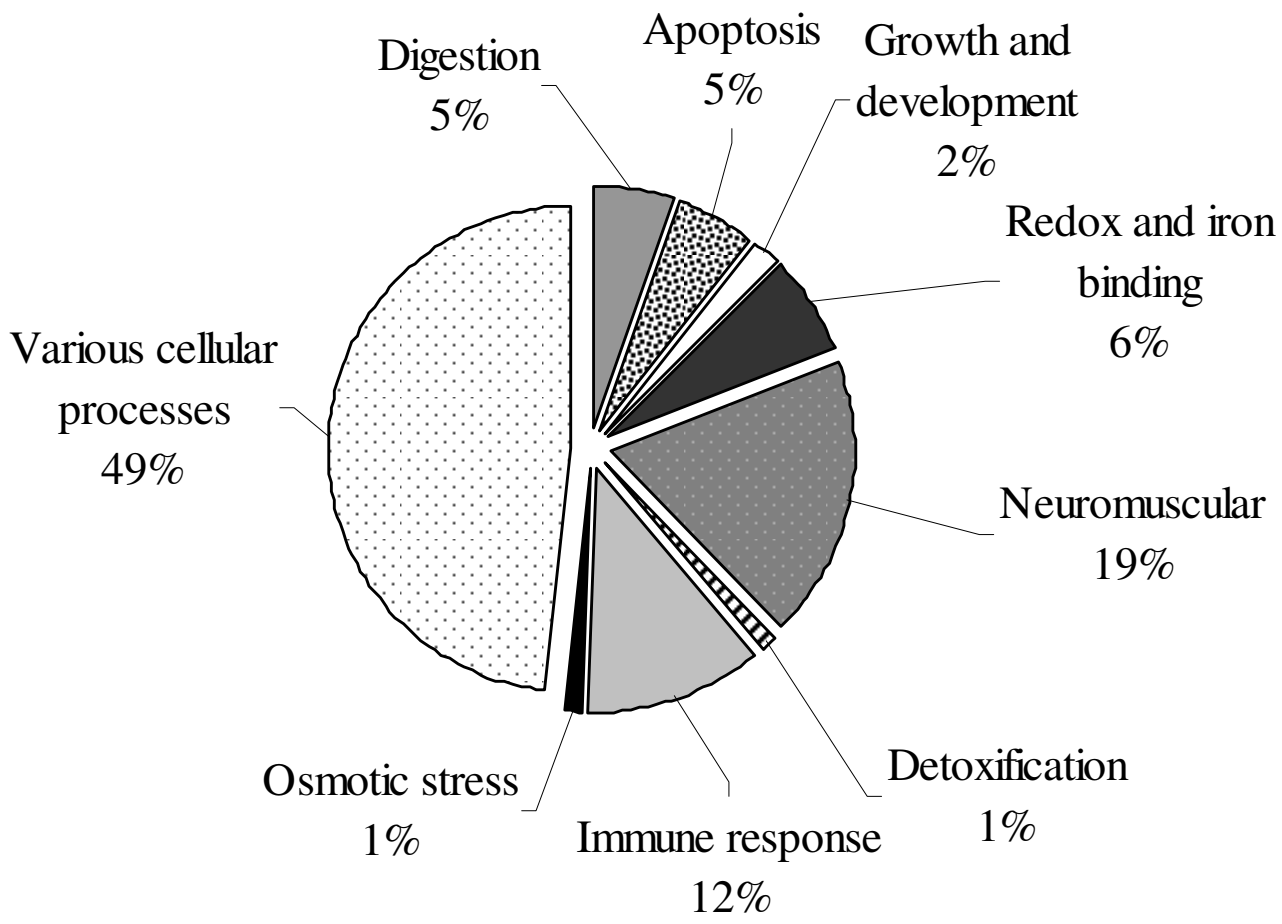

Figure 2
Functional classification of gene expression. Functional classification of genes responding to esfenvalerate exposure (0.0625 and 0.125 μg.L^{-1}) in 52-d old delta smelt. Percentages were calculated based on function and biological processes of 94 unique differentially expressed genes.

sented by genes coding for pancreatic enzymes, a zona pellucida protein; inferred to be choriogenin, and a growth hormone.

A selected group of genes, highlighted in bold in the gene list in table 1, were validated using q-PCR and further investigated for use as molecular biomarkers. These results are described below, in conjunction with differentially expressed genes identified through microarray application.

Specific effects of esfenvalerate exposure on gene expression

Pyrethroid insecticides are known sodium and potassium channel modulators [25], with axon demyelinating effects [27]. This the proof-of-principle microarray assessment of esfenvalerate exposure of 52-d old delta smelt, was successfully used to screen for, and further understand its mode of action, identifying neuromuscular responses, that were confirmed as highly significant through qPCR, corroborating known effects of this pesticide, but also pointing at other significant effects upon growth and development, digestion and the immune system. Gene expression assessed by qPCR on 10-d old delta smelt that verified these genomic responses are presented below. Correlations in expression between q-PCR investigated genes are shown in table 2 and fold changes in gene expression are summarized in figure 3 and table 3.

i. Neuromuscular responses

Parvalbumin expression in 10 d old larvae was induced 1.8-fold (t-test, $p = 0.008$) at 0.0313 μg.L^{-1} esfenvalerate, reducing in expression at higher concentrations. Localized in fast-contracting muscles, some endocrine tissues, in the

Table 2: Pairwise correlations of gene expression in esfenvalerate exposed 10-d old delta smelt.

	Aspartoacylase	Titin a	Microglobulin	Caspase-3	Parvalbumin	Hemopexin	ZPA	Myozenin	Creatine Kinase	PXR
Aspartoacylase	1 (0)									
Titin a	0.490 (0.402)	1 (0)								
Microglobulin	0.576 (0.310)	0.954 (**0.012**)	1 (0)							
Caspase-3	0.344 (0.571)	0.919 (**0.027**)	0.921 (**0.026**)	1 (0)						
Parvalbumin	0.284 (0.644)	0.953 (**0.012**)	0.928 (**0.023**)	0.981 (**0.003**)	1 (0)					
Hemopexin	0.629 (0.256)	0.819 (0.090)	0.951 (0.013)	0.860 (0.061)	0.822 (0.088)	1 (0)				
ZPA	-0.017 (0.979)	0.788 (0.113)	0.646 (0.239)	0.853 (0.066)	0.871 (0.054)	0.472 (0.422)	1 (0)			
Myozenin	0.309 (0.613)	0.898 (**0.038**)	0.949 (**0.014**)	0.909 (**0.033**)	0.943 (**0.016**)	0.892 (**0.042**)	0.677 (0.210)	1 (0)		
Creatine Kinase	0.255 (0.679)	0.916 (**0.029**)	0.865 (0.058)	0.984 (**0.003**)	0.978 (**0.004**)	0.759 (0.137)	0.932 (**0.021**)	0.859 (0.062)	1 (0)	
PXR	0.880 **(0.049)**	0.087 (0.890)	0.208 (0.737)	-0.113 (0.857)	-0.153 (0.805)	0.307 (0.616)	-0.475 (0.418)	-0.038 (0.952)	-0.218 (0.725)	1 (0)

Numbers represent correlation coefficients; r, and significance probabilities; (p), between ten selected biomarkers assessed with quantitative PCR. Bold p-values highlight significant correlations.

nervous system in GABAergic interneurons and in the brain [33], parvalbumin removes calcium from myofibrils, protecting neurons from hyper-excitability and facilitating muscle relaxation [34]. Accumulation of calcium in muscular tissue contributes to muscle degradation, muscular dystrophy and muscle fiber necrosis [35]. Estrogen is required for parvalbumin expression, thus estrogen receptor-β co-expresses with parvalbumin [36]. Estrogen is also required in brain development and has a protective neurological role, by regulating the activity of GABAergic systems within the hippocampus, basal forebrain and hypothalamus [37]. Differential expression of parvalbumin on exposure to esfenvalerate may be resultant of estrogenic effects. Pyrethroid pesticides have steroid receptor-binding activity [38] linked with endocrine disruption [39], thus exposure is likely to affect the population dynamics of wildlife not only through neuromuscular impairments, but also by affecting reproductive output [29,40]. Parvalbumin, could therefore, be a good indicator of possible endocrine-disruption as well as neuromuscular impairments.

Table 3: Mean and standard deviations in fold changes in gene expression of ten selected biomarkers in esfenvalerate exposed, 10-d old, delta smelt, assessed by quantitative PCR.

Gene\Concentration		0.000	0.031	0.063	0.125	0.250
Aspartoacylase	Mean	1.000	0.629	0.346	0.299	0.244
	SE	*0.240*	*0.100*	*0.078*	*0.035*	*0.088*
Titin	Mean	1.000	1.515	0.909	0.933	0.475
	SE	*0.529*	*0.198*	*0.095*	*0.192*	*0.096*
Microglobulin	Mean	1.000	1.420	0.760	0.828	0.628
	SE	*0.404*	*0.127*	*0.104*	*0.154*	*0.221*
Caspase	Mean	1.000	2.024	1.136	0.818	0.670
	SE	*0.336*	*0.432*	*0.043*	*0.117*	*0.094*
Parvalbumin	Mean	1.000	1.718	1.097	1.037	0.771
	SE	*0.241*	*0.151*	*0.125*	*0.062*	*0.107*
Hemopexin	Mean	1.000	1.501	0.521	0.548	0.612
	SE	*0.089*	*0.296*	*0.051*	*0.147*	*0.295*
ZPA	Mean	1.000	1.612	1.455	1.165	0.912
	SE	*0.415*	*0.321*	*0.224*	*0.210*	*0.574*
Myozenin	Mean	1.000	1.730	0.857	1.069	0.835
	SE	*0.212*	*0.205*	*0.121*	*0.093*	*0.188*
Creatine Kinase	Mean	1.000	1.799	1.265	0.968	0.750
	SE	*0.270*	*0.249*	*0.272*	*0.162*	*0.145*
PXR	Mean	1.000	0.737	0.668	0.737	0.729
	SE	*0.126*	*0.083*	*0.124*	*0.080*	*0.178*

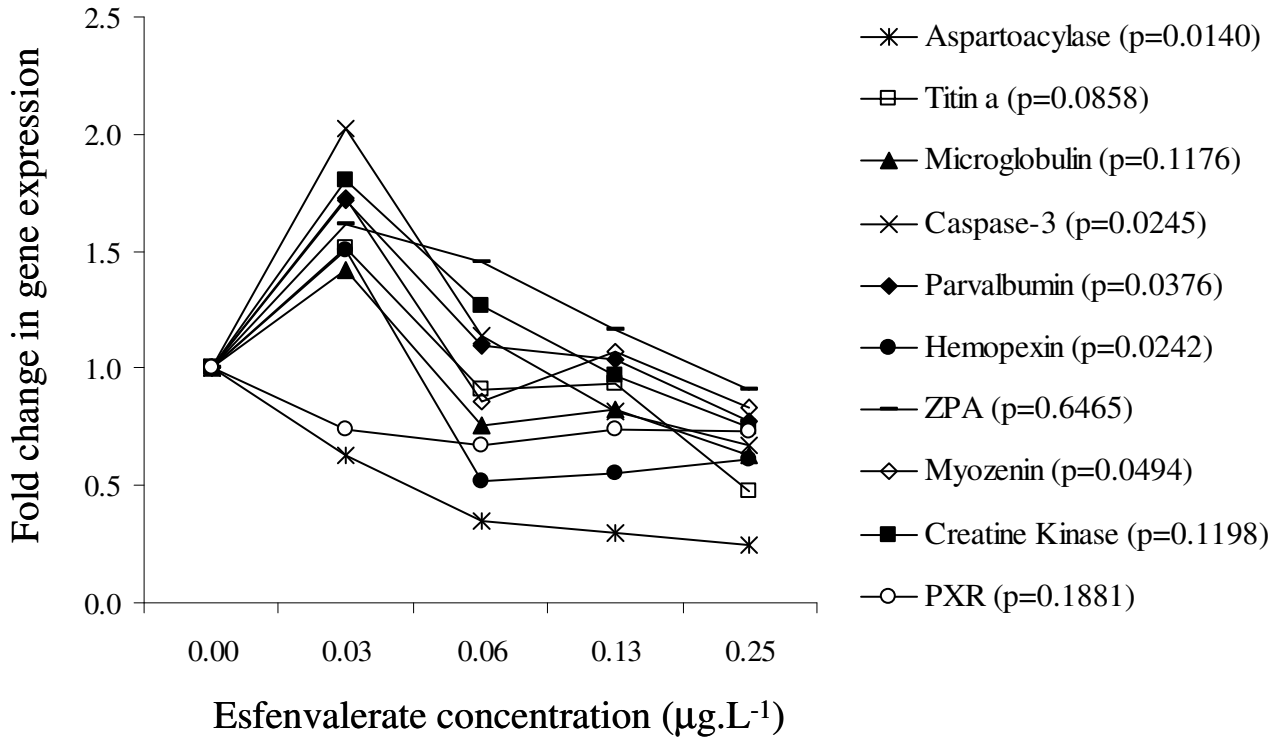

Figure 3
Molecular biomarkers responses. Fold changes in gene expression of ten selected biomarkers in esfenvalerate exposed, 10-d old, delta smelt, assessed by quantitative PCR. Significance in expression differences, as determined by One-way ANOVA, is shown in brackets in legend.

Interestingly, expression of aspartoacylase (ASPA) in exposed 10-d old delta smelt larvae was significantly affected at all concentrations, downregulating with increasing esfenvalerate concentration in a dose response manner, and correlating significantly with swimming anomaly at 24 h ($r = 0.913$, $p = 0.029$). Aspartoacylase catalyzes hydrolysis of *N*-acetyl-L-aspartate (NAA) to aspartate and acetate in the vertebrate brain [41]. Variations in NAA measured in urine, blood and brain, have been used as diagnosis of nervous system diseases such as Alzheimer's and multiple sclerosis [42,43]. Measurements of NAA, along with ADP levels determined by creatine kinase activity, are used to evaluate the energetic state of the brain, a positive linear correlation existing between NAA and ADP synthesis [44]. Deficiency in ASPA activity leads to degeneration of the myelin; an ensheathment that isolates and controls axonal activity, it is associated with schizophrenia [45], and is the established cause of leukodystrophy in Canavan's disease [43]. Abnormal myelination is known to result from acyltransferase deficiency [46]. Ependymin, a myelin associated glycoprotein related to memory formation and involved in neuronal regeneration [47], was also negatively affected by esfenvalerate. Myelin has been postulated as a probable modulator of ASPA activity [48] further affecting this critical pathway of neurological function. ASPA protein activity is a strong biomarker of brain damage and neurological impairment investigation, used regularly in human and veterinary disease diagnostics [49].

Creatine kinase was significantly up-regulated at 0.0313 $\mu g.L^{-1}$ esfenvalerate (t-test, $p < 0.05$). Creatine kinase protein is used not only as a diagnosis of brain energetic value as mentioned above, but also of diseases like cardiac infarction and skeletal muscle necrosis [50]. In muscle, creatine kinase is specifically bound to sarcoendoplasmic reticulum, and regulates calcium uptake and ATP/ADP ratios [51], thus is directly involved in muscle contraction. Of interest here, are the pathway links and correlating responses ($r = 0.98$) between parvalbumin; facilitating muscle relaxation by binding calcium, and creatine kinase, which regulates calcium uptake. These two param-

eters on their own indicate muscular activity impairments, creating strong links with observed larval swimming behavior.

Titin expression also correlated significantly with parvalbumin (r = 0.95) and creatine kinase (r = 0.92), though not statistically significant in q-PCR assessments of 10-d old smelt larvae. Titin is an important protein also involved in muscle contraction, responsible for muscle elasticity and is the molecular scaffold for thick actin filament formation, forming a connection between filaments and the muscle Z-line [52]. Myozenin, another protein involved in muscle contraction, was significantly influenced by esfenvalerate exposure (t-test, $p < 0.05$) in 10-d old larvae. Co-regulating with Titin (r = 0.90), myozenin is a Z-line, α-actinin- and γ-filamin-binding protein expressed predominantly in skeletal muscle, and has been suggested as a biomarker for muscular dystrophy and other neuromuscular disorders [53].

ii Immune responses

In this study we have identified a significant alteration in expression of several genes involved in immune responses, most of them with links to neurological damage. β-microglobulin, a small protein normally found on the surface of many cells, including lymphocytes, is known to be involved in cell protection [54]. β-microglobulin is almost exclusively catabolized in the kidney and its excretion is an indication of long term nephrotoxicity [55]. High concentrations of β-microglobulin are reported to inhibit generation of functional dendritic cells [56], thus an increased amount in the blood or urine may be a sign of neural degeneration and of certain diseases, including some types of cancer, such as multiple myeloma or lymphoma. β-microglobulin levels are also reported to rise following viral infection and its reduced expression can compromise the immune system [57]. Interestingly, exposure to esfenvalerate resulted in a significant increase of pathogen susceptibility in chinook salmon [9]. β-microglobulin assessment with q-PCR did not show any significance in expression in 10 d old larvae (t-test, $p = 0.131$), however, an overall increase was observed at low concentrations of esfenvalerate, correlating significantly with expression of other genes investigated, further discussed below.

Multiple sclerosis is caused by an immunological attack on myelin [58], decreasing NAA and resulting in neurological instability. Furthermore, oxidative stress is inductive of apoptosis of myelin-reactive T cells [59]. A putative T cell receptor gene was identified through microarray screening, probably reacting to pyrethroid exposure, acting upon the myelin sheath and causing further neurological damage and cell death.

iii Apoptotic responses

Caspases (*c*ysteine-*asp*artic acid prote*ase*) belong to a family of cysteine proteases that cleave other proteins, such as the precursor forms of the inflammatory cytokines interleukin 1-β and interleukin 18, into active mature peptides and are also involved in programmed cell death; or apoptosis [60]. Enzymatic activity requires an aspartic acid residue, and plays a critical role in the regulation of proinflammatory cytokines [61] that are associated with septic shock and autoimmune syndromes [62]. Upregulation of proinflammatory cytokines were reported in viral infected salmon, which further increased in expression following esfenvalerate exposure [63]. Caspases contribute to the pathogenesis of neurodegenerative disorders such as ischemia, Krabbes and Huntington's diseases, Alzheimer's, and other leukodystrophic diseases resulting in neural degeneration [64,65]. Moreover, caspase inhibitors have been suggested as therapeutic treatments for neurodegenerative diseases [66]. Low concentrations of esfenvalerate; 0.0313 $\mu g.L^{-1}$, significantly induced caspase-3 expression 2-fold (t-test, $p = 0.002$) in 10-d old delta smelt. As caspases are activated by aspartic acid, induction may be suggestive of increases in substrate residues, along with inflammatory cytokines, probable effects upon the immune system, and subsequent neurodegeneration. Furthermore, a decrease in ASPA expression could be suggestive of reduced breakdown of NNA to aspartate and acetate, required as substrate for caspase activity, and synthesis of proteins required in repair mechanisms.

iv. Redox and metal ion binding

Upregulation of hemopexin was confirmed by quantitative PCR in 10-d old delta smelt exposed to 0.0313 $\mu g.L^{-1}$ esfenvalerate. Hemopexin-like protein, a gene sequence displaying vast similarities to warm-temperature-acclimation-related-65 protein (WAP65) on BLAST homologies with Japanese medaka (*Oryzias latipes*), was identified as significantly upregulated through microarray screening. Hemopexin is synthesized by Schwann cells following nerve injury [67], accumulation has been reported in the peripheral nervous system following axonal lesions, and is specifically regulated during repair, returning to normal levels on axonal regeneration [68]. Wallerian degeneration occurs after axonal injury and is critical for repair, it is characterized by axonal and myelin degeneration [69], is accompanied by macrophage invasion and subsequent synthesis of hemopexin [67]. Hemopexin appears to play a significant role in neural regeneration, but may be resultant of oxidative stress mediated T cell activity on myelin sheaths (described above, under immune responses). Though upregulation follows nerve injury and there are strong connections with apoptosis, we classify hemopexin under oxidative stress as it has a high affinity with heme and reportedly plays a strong role in both heme transport and preventing heme-catalyzed oxidative

damage [70]. Moreover, pyrethroids have been shown to generate free radicals and induce oxidative stress [71]. Heme is known to respond to nerve injury, and has been suggested to play a role in neurodegenerative disorders [72], and hemopexin-mediated heme transport was reported to significantly decrease levels of transferrin receptor mRNA in HeLa cells [70]. Transferrin was also identified by microarray screening as significantly upregulated by esfenvalerate exposure. The primary role of transferrin is the delivery of iron across the blood brain barrier, and its expression in brain is not only related to myelin production, but may be a permissive agent in the process of myelination [73]. Furthermore, hemopexin and transferrin reportedly act by similar receptor-mediated mechanisms [74].

v. Growth and development

Microarray analyses identified a gene with high homology to egg envelope glycoproteins within the zona pellucida (ZPA) referred to as choriogenins, in fish [75]. This was significantly expressed in 52-d old larvae, however, no statistical differences in expression of ZPA were measured with qPCR in 10-d old larvae exposed to esfenvalerate. Choriogenin is reportedly more sensitive to endocrine disrupting chemicals (EDCs) than estrogen receptors and vitellogenin [76]. Composed primarily of glycoproteins with various functions during fertilization and development, choriogenin has been suggested as a biomarker of exposure to endocrine disrupting chemicals, as it is induced in late stage embryos, larvae and adult male fish exposed to estrogens [76,77]. Choriogenin is synthesized in liver of adult females, in response to estrogen, transported in blood and incorporated into the fish egg envelope; chorion or zona pellucida (ZPA), an extracellular matrix that surrounds the oocyte and early embryo [78]. Expression was notably elevated at low pyrethroid concentration, and it responded in a similar fashion to creatine kinase ($r = 0.93$), though no significant links were identified between these two biomarkers.

vi. Detoxification

Pregnane × receptor (PXR), involved in the detection of toxic substances and a key regulator of xenobiotic metabolism, was identified through microarray assessments, as downregulated in 52-d old larvae. PXR is a steroid receptor and transcriptional regulator of detoxification mechanisms such as cytochrome oxidases, and phase II conjugating enzymes such as glutathione-*S*-tranferases [79,80]. Downregulation of PXR expression has been linked with growth inhibition and cell death in rats and human cell lines following exposure to medroxyprogesterone and estradiol, known PXR ligands [80], further supporting identified apoptotic responses, steroid receptor-binding and endocrine disruption activity of esfenvalerate [38,39]. PXR expression was not significantly different in q-PCR assessments of 10-d old larvae exposed to esfenvalerate, however, overall expression declined in a dose response manner, correlating with ASPA ($r = 0.880$; $p = 0.049$), making it a notable candidate of xenobiotic detection for future biomarker investigations in the delta smelt.

vii Osmotic Stress

A hyperosmotic glycine rich protein was identified with the microarrays as significantly downregulated by exposure to 0.0625 $\mu g.L^{-1}$ esfenvalerate in 52-d old larvae. Osmoregulation is physiologically controlled by chemical messages from the endocrine system, along with cell signalling and nerve transmission [81]. Pyrethroids have been suggested to induce osmotic imbalances in common carp larvae [82] which are linked to effects on ATPase activity responsible for maintaining the Sodium transmembrane electrochemical gradient [83]. Larval fish are under direct exposure to osmotic stress as their endocrine system is not fully developed [84]. Parvalbumin and choriogenin expression have indicated possible effects on the endocrine system, thus expression of this hyperosmotic glycine rich protein may be directly caused by conditions affecting endocrine regulation.

viii Digestion

Chitinase was identified through microarray analysis as being downregulated by esfenvalerate exposure. Chitinase is the principal enzyme involved in digesting chitin, a major component of insect and crustacean exoskeleton [85]. Larval smelt were fed on artemia during the pre-exposure acclimation period. Not only chitinase but, other digestive enzymes; apolipoproteins, pancreatic enzymes, carboxypeptidase precursors and chemotrypsinogen, were also significantly upregulated following exposure in 52-d old larvae. Effects on digestion alone will undoubtedly have significant effects on growth, which when combined with hypothesized feeding reduction resulting from impaired swimming would lead to significant malnutrition. Contaminants affect a whole ecosystem, at all levels, and dramatic reductions in copepods, cladoceran and amphipod populations; organisms predated upon by the delta smelt, have been reported in the Sacramento delta [86]. Scarcity in food and reduced ingestion ability, besides digestion will significantly affect population dynamics of any specie.

Conclusions

Microarray technology was used as an initial screening of probable genes responding to esfenvalerate exposure therefore no multiple testing correction was applied. We have, however, examined and confirmed effect of esfenvalerate upon some of the genes in a different age group of larval delta smelt, identifying significant responses that are primarily linked with swimming behavior. Some

responding genes can be classified within different functional groups. Due to the measured behavioral responses, the classification approach contains a certain bias towards understanding neuromuscular effects. It is interesting that qPCR measurements have identified a greater response at the lower concentrations, implying homeostatic alterations, at environmentally relevant concentrations. Most genes did not display a desired dose response correlation associated with usable biomarkers, but did support responses within the suite of genes investigated, somewhat validating their use within a broader biomarker approach. Hemopexin for example is known to be involved in axon repair, and the myelin sheath surrounding the axon needs to be degraded for this repair to be processed, hypothesizing therefore that ASPA downregulation is resultant of neurological damage. The subsequent decrease of hemopexin expression at higher exposure concentrations, and further decrease in ASPA, may be indicative of repair impairments.

What becomes apparent from this study is that exposure to sublethal concentrations of esfenvalerate results in neurological damage and a series of compensatory molecular responses that attempt to repair nerve damage. We would hypothesize that induction of transcription of the genes encoding ASPA, hemopexin, parvalbumin and creatine kinase are part of a pathway of damage triggered repair mechanisms, responding to esfenvalerate insult. Reduction in expression of ASPA indicates that myelin sheaths may be degraded, resulting in a number of detrimental effects on the lesion sites, and similarly, muscular structure and function is being compromised as measured by alterations in titin and myozenin expression. The expression of β-microglobulins could be a compensatory reaction to toxic damage, protecting cells from infections in a susceptible immune system caused by exposure to esfenvalerate. Previous studies, carried out in esfenvalerate exposed chinook salmon have reported a compromised immunity and significantly higher susceptibility to infection [9,63]. This is particularly important in younger organisms that are generally more susceptible than adults. Furthermore, polluted waters not only contain mixtures of contaminants, but also harbor multiple pathogens that will further affect health parameters.

Behavioral endpoints, such as swimming behavior, are amongst the most sensitive and ecologically relevant parameters to assess sublethal toxicity of neurotoxic chemicals [29]. The high susceptibility of delta smelt to esfenvalerate, mediated neurological damage resulting in impaired swimming ability, also raises questions on the likely effects upon their chemosensory system; olfactory system, important in sensing reproductive pheromones, mediating reproduction. Females synthesize sex hormones stimulating male reproductive behavior [87]. Neurological damage affecting the olfactory nerves, the brain and or entire nervous system, could lead to further impairments in reproductive success following exposure to pyrethroids. Damage to the olfactory system has been used as a sublethal toxicological endpoint in fish, in studies investigating behavior following pesticide exposures [88]. Pyrethroids are known to affect the olfactory system [89]. A chemosensory gene, ictacalcin, responding to esfenvalerate exposure was also differentially expressed on the microarray. Ictacalcin is a gene originally identified in catfish (*Ictalurus punctatus*), involved in chemosensory tissues, and highly expressed in barbell, olfactory mucosa and gill [90]. Differential expression of this gene may indicate that further behavioral parameters, not investigated in this study, such as recognition, alarm response, feeding, imprinting and homing, gamete release and synchronization, contaminant avoidance [88], and other behavioral parameters that are governed by chemosensory system, could be compromised. We could speculate that outside laboratory conditions, neuromuscular and chemosensory impairments would probably result in higher ecological parameters being affected through inability to swim against water currents, making them more susceptible to predation and reducing their ability to obtain food. Furthermore, effects on chemosensory parameters would lead to migratory, reproductive, predator and contaminant avoidance impairments.

Inhibition of repair mechanisms, leading to neuromuscular damage and eventual death, was behaviorally observed throughout exposure, as impairment in swimming ability. The ability to use molecular biomarkers of neuromuscular effect further strengthens links between mechanistic effects with parameters of ecological relevance. Our study supports the use of gene expression as a productive way of understanding modes of actions of individual chemicals in endangered species. Furthermore, this screening and interrogative approach permits the identification and development of biomarkers for species of concern in which prior information is limited and allows for investigations into problems specific to the organism in question; assessing possible causes of detrimental effects and resulting influences on individual performance and hypothesizing effects upon population dynamics. A suite of biomarkers developed in this manner, though additions and subtractions are required from the presented list, could be used to aid monitor impacts of stressors upon organisms within a specific environment and could be an essential tool in determining causative factors of population decline in the delta smelt and other threatened species. The selected biomarkers clearly need to be further investigated and validated against other known contaminants, and suitability in field applications.

Methods

Microarray construction

We constructed a delta smelt microarray using 8448 PCR amplified fragments from a normalized cDNA library. A cDNA library was created using expressed sequence tags (ESTs) ligated into *p-BS* plasmid vectors and cloned into chemically competent *Escherichia coli* cells (BioS&T Inc, Montreal, Quebec, Canada). RNA for library construction was obtained from a number of larval, juvenile and adult delta smelt, ranging from unexposed, control conditions, to fish from exposures to high temperature (25°C), and sublethal concentrations of copper, esfenvalerate, and a six field water samples from throughout the Sacramento-San Joaquin estuary. Products were PCR amplified from 1 μl bacterial suspension, and visualized on agarose gels. Purified PCR fragments ranging in size from 1-4 kb, along with control spots, were pin-printed in duplicate onto epoxysilane coated glass slides (Schott-Nexterion, USA) in a 20 × 19 block format, with 48 blocks per microarray. Microarrays were printed using a Lucidea Array Spotter (Amersham) at the Array Core facility at UC Davis (since closed down). 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 consisted of 1× Nexterion buffer solution.

Esfenvalerate exposures

Delta smelt larvae aged 10 d and 52 d were exposed for 24 h, in two separate experiments, to a range of esfenvalerate concentrations; 0.0313, 0.0625, 0.125, 0.250 and 0.500 $\mu g.L^{-1}$ (nominal) in laboratory control water, with corresponding laboratory and solvent controls. Concentrations were measured at the start of the experiment by the Water Pollution Control Laboratory at the Department of Fish and Game (Rancho Cordova, California, USA), only single measurements were taken per treatment (results not shown), therefore we present the data in terms of nominal concentrations. Laboratory control water consisted of deionized water amended to US EPA moderately hard standards (80-100 $mg.L^{-1}$ $CaCO_3$) and 200 μl/L methanol was used as solvent carrier. Salinity was adjusted with Instant Ocean salt to match hatchery rearing conditions (range 650 $\mu S.cm^{-1}$ to 900 $\mu S.cm^{-1}$).

Average wet weights of 10-d to 52-d old larval delta smelt ranged from 0.5 to 2.5 mg respectively. Larvae were obtained from the Fish Conservation and Culture Laboratory (FCCL) UC Davis, Byron, CA, transported in cool, oxygenated 2-gallon black buckets, and held overnight in the laboratory at 17°C and a 8 h:16 h D:L light cycle. The following day, ten larvae were transferred to each 2-L beaker containing 1 L of aerated control water or esfenvalerate treatment. Each treatment consisted of 4 replicates and tests were performed at 8 h:16 h D:L cycle, at a water temperature of 17°C ± 1.2°C. The pH during the tests was 7.1 - 7.5. Dissolved oxygen levels were within the acceptable range for delta smelt (above 6.5 $mg.L^{-1}$)[91]. Larvae were fed rotifers the day before the test start, but not during the 24 h exposure. Rotifer cultures were obtained from FCCL. During exposure, larvae were observed for aberrant swimming behavior, and surviving fish were scored after 4 h and 24 h. Swimming behavior was assessed by observing each tank for 5 min as described in Geist *et. al.* [17]. Any pronounced deviation (> 1 min) from normal (control) swimming patterns were recorded as abnormal. Effects on swimming performance (EC_{50}) and mortality (LC_{50}) were assessed using linear regression analysis with Environmental Toxicity Information System (CETIS) by Tidepool Scientific Software (McKinleyville, CA, USA).

Four surviving 52 d old larvae from solvent controls, and exposures to 0.0625 and 0.125 $\mu g.L^{-1}$ esfenvalerate, were used for microarray analyses, hybridized in a reference design against a pool of RNA from all treatments. Four replicate 10 d old larvae from each treatment (controls and 0.0312, 0.0625, 0.125 $\mu g.L^{-1}$ esfenvalerate) were used for biomarker analyses and gene expression verification using quantitative PCR (q-PCR).

All experiments and use of test organisms were approved by the UC Davis Institutional Animal Care and Use Committee (Animal Use Protocol for Animal Care and Use #13361). This institution is accredited by the Association for Assessment and Accreditation of Laboratory Animal Care, International (AAALAC) and has an Animal Welfare Assurance on file with the Office of Laboratory Animal Welfare (OLAW). The Assurance Number is A3433-01. The IACUC is constituted in accordance with U.S. Public Health Service (PHS) Animal Welfare Policy and includes a member of the public and a non-scientist.

RNA isolation, cDNA synthesis and fluorescence labeling

RNA was extracted from whole, individual organisms using a standard phenol:chloroform protocol with Trizol Reagent (Invitrogen). Fifteen micrograms of total RNA were used for 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). Each experimental sample and control was combined with a reference pool cDNA prior to hybridization using an automated Tecan HS4800 hybridization station. Slides were scanned using a GenePix 4000B scanner (Axon Instruments).

Microarray images and data from esfenvalerate exposed delta smelt can be accessed at http://www.vet

med.ucdavis.edu/apc/WernerLab/subpagpelagic_organism_decline.html; POD archive data.

Microarray Analyses

Normalization and analytical methods are described in Loguinov *et. al*. [32] and Smyth [92]. In brief, 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 α-outlier-generating model was used to identify differentially expressed genes by applying the following decision rule for multiple-slide data analysis: a given gene was selected as a candidate if it was detected as significantly up- or downregulated in 4 of 4 replicates (raw p-value = 0.0625 using exact binomial test and considering outcomes as Bernoulli trials). The approach did not use scale estimator for statistical inference and, therefore, it did not require between-slide normalization. This method however, detected only one significant differentially expressed candidate gene at the highest exposure concentration (0.125 $\mu g/L^{-1}$), (with no significant annotation identity - see results and discussion). As a result, a second analytical method was applied to increase the number of probable genes for consideration in biomarker development. Thus we further analyzed the data using LIMMA GUI (Linear model for microarray analysis graphical user interface) [92], written in the R-programming language available through Bioconductor http://www.Bioconductor.org. Data was normalized within arrays using print-tip Lowess and between arrays applying aquantile normalization methods [92]. A linear model fit was computed using the duplicates on the arrays and the least-squares method, no multiple assessment methods were applied to eliminate false positives as our aim was to increase the number of genes available for biomarker assessment, and qualify these through quantitative PCR.

Sequencing and Annotation

Sequencing was carried out at the CA&ES Genomic Facility, UC Davis. Basic Local Alignment Search Tool; translated nucleotide (BLASTx) searches were performed on specific fragments that responded significantly to the exposure treatments. Only genes that were differentially expressed following esfenvalerate exposure were sequenced. Sequences were annotated according to homologies to protein database searches using translated nucleotide sequences and direct nucleotide queries http://blast.ncbi.nlm.nih.gov/Blast.cgi. Sequences were only annotated if they were found to have a BLASTx match with the expect value smaller than 1×10^{-5} and a score above 50.

Functional Classifications

Differentially expressed genes were classified according to gene ontology http://www.uniprot.org/uniprot, and information gathered from literature, into functional groups. Classification was carried out based on gene expression changes in respect of control subjects, regardless of whether these were up or downregulated, or exposure concentrations. Specific genes of interest were selected for further investigation using quantitative PCR.

Table 4: Primers and probes used for molecular biomarkers: Primer pairs and TaqMan probes used in q-PCR assessments.

Accession no.	Gene		Primer Sequences	Probe No.
FJ711577	Aspartoacylase	Left:	ggaggcacacatgggaatg	109
		Right:	cttcctctgaatctctgttccattatc	
FJ711576	Parvalbumin	Left:	gaccaagacaagagtggcttca	101
		Right:	tctggcaccagcagagaagtt	
FJ711580	β-2 microglobulin	Left:	tctttcgcggtcatctttctc	22
		Right:	ggttgtggccatacacctgaa	
FJ711579	Hemopexin	Left:	catgcactacgaggacgacaag	143
		Right:	tggtagtagctgaacaccttgctg	
FJ711578	Caspase-3	Left:	gagaaccggtatgaaccaacg	159
		Right:	tccaagcttcccaaacactttc	
FJ711575	Titin a	Left:	tgatcactggcgtgaaagagg	159
		Right:	caagctcattggacagtttgagg	
FJ711581	ZPA	Left:	catgcggctgagtttggataa	106
		Right:	tgccattgatagcatcaacttca	
FJ711583	Myozenin*	Left:	ccaatgtcgtgctggtacacc	106
		Right:	ctgccagacattgatgtagcca	
FJ711584	Creatine kinase*	Left:	cgatcggcgttggagatg	163
		Right:	gccaagttcaacgagattctgg	
FJ711582	PXR	Left:	tgaggcggtggagaagag	144
		Right:	gaggcggtggagaagag	

* indicates biomarkers obtained from preliminary studies carried out during microarray development.

Biomarker development

Genes were selected according to level of expression significance, knowledge base from literature, and functional classification. Primer and probes for qPCR analyses were designed using Roche Universal Probe Library Assay Design Center https://www.roche-applied-science.com. Designed primers were obtained from Eurofins MWG Operon http://www.eurofinsdna.com, and TaqMan probes were supplied by Roche. Sequences for all genes assessed by qPCR analyses have been submitted to genbank http://www.ncbi.nlm.nih.gov. Primers and probes, and genbank accession numbers, for investigated biomarkers are detailed in table 4. Myozenin and creatine kinase, though not resulting from the current study, were genes identified in investigations carried out during microarray development, and added to the selected biomarkers due to our interest in neuromuscular activity.

Quantitative PCR

A total of 1.5 μg RNA was cDNA synthesized using random primers, and diluted to a total of 50 μl with nuclease free water to generate sufficient template for qPCR analysis. TaqMan Universal PCR Mastermix (Applied Biosystems) was used in q-PCR amplifications in a reaction containing 10mMTris-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 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 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.

Quantitative PCR data was analyzed using the relative quantification 2(-Delta Delta CT) method ()([93]. In the absence of house keeping genes, expression was calculated relative to the mean of controls in respective exposures. Surviving larvae from each replicate of the 10-d old exposed delta smelt were used for q-PCR analyses. One-way ANOVA was used to assess differences in gene expression through out the exposure concentrations, and data were further assessed using Student's T-test at individual concentrations in respect to solvent controls.

Abbreviations

EST: expressed sequence tag; q-PCR: real-time quantitative polymerase chain reaction; ASPA: aspartoacylase; ESA: endangered species act; CESA: California endangered species act; LIMMA GUI: linear model for microarray analysis, graphical user interface; BLAST: basic local alignment search tool; GABA: gamma-aminobutyric acid; NAA: *N*-acetyl-L-aspartate; ADP: adenosine diphosphate; WAP65: warm-temperature-acclimation-related-65 protein; EDC: endocrine disrupting chemical; ZPA: zona pellucida; PXR: pregnane × receptor; p-BS: p-Bluescript; IPTG: isopropyl β-galactosidase; LB: Luria Bertani; FCCL: Fish Conservation and Culture Laboratory; UNG: Uracil N-glycosylase; CT: cycle threshold.

Authors' contributions

All authors were involved in designing the experiments. JG carried out esfenvalerate exposures recording swimming abnormality and mortality. REC designed, developed and applied the microarrays with assistance from JP and HW. REC and JP prepared, hybridized and scanned microarrays. REC and AVL performed microarray analyses. REC and LSD designed qPCR assays and performed respective assessments and analysis. REC drafted the manuscript and all authors read, contributed intellectually and approved the final manuscript.

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Linking molecular biomarkers with higher level condition indicators to identify effects of copper exposures on the endangered delta smelt (*Hypomesus transpacificus*).

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Linking molecular biomarkers with higher level condition indicators to identify effects of copper exposures on the endangered delta smelt (*Hypomesus transpacificus*).

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Abstract:

The experimental combination of molecular biomarkers with higher level condition indicators has allowed for interpretation of responses in a functional context that can be used to predict detrimental outcomes caused by contaminant exposure. In fish, not only swimming behavior, but maintenance of optimal swimming performance is of particular importance for optimal fitness. The delta smelt (*Hypomesus transpacificus*) is an endangered pelagic fish species endemic to the Sacramento-San Joaquin estuary, California, and considered an indicator of ecosystem health. Copper is a contaminant of concern in Californian waterways. Functional classifications of microarray responses are presented along with complementary quantitative PCR in measuring effects upon neuromuscular, digestive and immune responses in delta smelt exposed to copper. Differences in sensitivity were measured between juvenile and larval delta smelt ($LC_{50\text{-}96h}$= 25.2 and 80.4 $\mu g.Cu^{2+}.L^{-1}$ respectively). Swimming velocity of declined on exposure in a dose dependent manner. Genes encoding for aspartoacylase, hemopexin, α-actin and calcium regulation proteins were significantly affected by exposure and were functionally linked with measured swimming responses. Effects on digestion were measured by upregulation of chitinase and downregulation of amylase. Downregulation of tumor necrosis factor indicated a compromised immune system. Results from this study, and many others support the use of functionally characterized molecular biomarkers to assess effects of contaminants in field scenarios. We thus propose that in order to attribute environmental relevance to molecular biomarkers, research should concentrate on their application in field studies with the aim of developing monitoring programs.

Keywords: 'Hypomesus transpacificus', 'delta smelt', microarray, biomarker, copper, "stress response", "aquatic ecosystems", endangered

Introduction.

The application and significance of biomarkers in an environmental context has been criticized due to the lack of linkage between biomarker response in individual organisms and effects (such as reproductive fitness or population decline) at higher levels of organization (e.g., populations and ecosystems) [1-8]. The experimental combination of biomarkers with indicators of population or ecosystem condition, allows for the evaluation of effects upon an individual and subsequent extrapolation of endpoints such as population effects. Recent studies have hypothesized, identified and demonstrated links between gene expression and higher levels of organization [9-16]. The success behind the use of biomarkers as early and sensitive warning tools thus lies on interpreting biomarker responses in individuals in the context of affected cellular pathways, integrated with extensive life history knowledge of the species in question. This requirement is especially true when assessing effects on non-model species, or organisms living in ecological systems where sensitivity to stressors could greatly differ from sentinel organisms.

In fish, not only swimming behavior, but maintenance of optimal swimming performance is of particular importance for optimal fitness. A number of life history variables are dependent on swimming ability, including respiration, feeding, predator prey interactions, and social interactions such as courtship and spawning, which are fundamental to survival, growth and reproduction, the most important traits in evolutionary success. Contaminant exposures that predominantly affect neuromuscular structure and activity will undoubtedly translate to swimming impairments, however, other maintenance aspects, such as immune system and acquisition of adequate nutrients, will also play a role. Furthermore, exposure may affect olfactory senses and related behavioral responses, like contaminant avoidance and homing that will further affect individuals' chances of survival and reproduction, and as such have direct effects on population dynamics. Contaminant avoidance is, in itself, generally seen as a beneficial response, however, should the avoidance coincide with homing and identification of limited spawning sites, would impinge on reproduction and consequently population dynamics [17].

The delta smelt (*Hypomesus transpacificus*) is an endangered pelagic fish species endemic to the Sacramento-San Joaquin estuary, California, whose abundance has dramatically declined since the 1980s, and more precipitously in recent years [18-21]. A number of complex factors, such as freshwater export and habitat destruction [22, 23], have been attributed to the decline of delta smelt in its native environment, with contaminants being another key issue. A more recent steep decline has prompted considerable efforts to understand the causative factors of reduced population [21, 24], especially because several other pelagic species have shown similar population trends.

Delta smelt are restricted to the Sacramento-San Joaquin estuary, spawning in late winter and early spring, at limited freshwater sites both known and speculated upon, based on sediment type and other physicochemical properties such as slope, vegetation, depth, temperature and salinity [25, 26]. A major site of delta smelt spawning is in the Lower Sacramento River from which hatched larvae are transported with the flow to brackish waters down-stream, where they mature, during a short-lived one-year cycle [27, 28]. This spawning site is located within agricultural areas and down-stream from the Sacramento

municipal wastewater treatment plant. Thus, emerging larvae are potentially exposed to a cocktail of contaminants, coinciding with early pest control chemical applications that mix with urban effluent contaminants.

Copper is a contaminant of concern in Californian waterways. It is not only common in urban storm-water runoff, and transport off old mining sites, but it is regularly used as a pesticide and fungicide in agricultural areas. Seasonally fluctuating dissolved copper concentrations in the Sacramento River have been reported approximating 2 $\mu g.Cu^{2+}.L^{-1}$, however, concentrations in tributaries (e.g. Arcade Creek) have been measured above 6 $\mu g.L^{-1}$ [29], and concentrations have been reported to exceed 500 $\mu g.L^{-1}$ in rice field effluents following application [30]. The delta smelt have been reported to be highly sensitive to copper [31] and its extensive application in Californian agriculture likely affects ecosystem health.

Copper, though essential for multiple cellular proteins, can be toxic to many aquatic organisms including fish. The mode of action of copper in several fish species has been reported to involve inhibition of Na^+ channels in gill epithelium although other mechanisms are likely to be important as well [32]. Our knowledge on the effects of copper on model organisms is extensive, making this an ideal contaminant to utilize in this biomarker assessment proof-of-concept study, where our aim is to link molecular responses with higher level condition indicators, in this study, swimming performance. We describe in this paper, the effects of copper upon delta smelt at sensitive larval and juvenile developmental stages, the development of molecular biomarkers and their link with swimming performance. We emphasize the need for inclusion of molecular biomarkers in monitoring programs in order to understand mechanisms by which contaminants exert effects upon endangered organisms.

Methods

Fish exposures and swimming assessments

Delta smelt were obtained from the Fish Conservation and Culture Laboratory, UC Davis and maintained for a minimum of 24 hours in experimental conditions prior to test initiation. Two separate tests were conducted with juveniles and larvae, and used to assess gene expression through microarray and quantitative PCR (qPCR) applications respectively. Swimming behavior of larval fish was investigated and compared to q-PCR responses as detailed below. All experiments and use of test organisms were approved by the UC Davis Institutional Animal Care and Use Committee (Animal Use Protocol for Animal Care and Use #13361). This institution is accredited by the Association for Assessment and Accreditation of Laboratory Animal Care, International (AAALAC) and has an Animal Welfare Assurance on file with the Office of Laboratory Animal Welfare (OLAW). The Assurance Number is A3433-01. The IACUC is constituted in accordance with U.S. Public Health Service (PHS) Animal Welfare Policy and includes a member of the public and a non-scientist.

i. Exposures used for microarray analysis:

Juvenile delta smelt (90-d old) were exposed for 7 days to 4.2, 8.4, 21.0 and 42.0 $\mu g.C^{2+}.L^{-1}$; sourced from $CuCl_2$ (dihydrate). Controls were maintained in diluted well-water adjusted to a specific conductance (SC) of 450 $\mu S.cm^{-1}$ using Instant Ocean, and a pH of 8.45, using HCl. Juveniles were acclimated to control water for 24 h prior to test initiation. Replicate experimental treatments (n=4) were initiated with 10, 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% of the water in each replicate was renewed at test initiation and on days 2, 4, and 6. At test end, surviving fish were euthanized with MS-222 (tricaine methanesulfonate, Sigma, St. Louis, MO, USA), then measured, weighed and snap-frozen in liquid nitrogen and stored at –80°C for subsequent analyses. Surviving juveniles from 42.0 $\mu g.C^{2+}.L^{-1}$ were assessed against controls, utilizing the developed microarrays (see below).

ii. Exposures used for quantitative PCR and swimming ability analyses:

Larval delta smelt (47-d old) were exposed for 4 days to 24.0, 52.4, 105.0 and 213.0 $\mu g.C^{2+}.L^{-1}$; sourced from $CuCl_2$ (dihydrate). Controls were maintained in hatchery water with specific conductance (SC) of 930 $\mu S.cm^{-1}$ and pH of 7.9. Larvae were acclimated to control water for 24 h prior to test initiation. Antibiotics, (Maracyn and Maracyn-2, Virbac AH Inc., Fort Worth TX) were added during the acclimation period at concentrations of 5.3 $mg.L^{-1}$ Maracyn (erythromycin) and 0.26 $mg.L^{-1}$ Maracyn-2 (minocycline), to eliminate any gram-positive and gram-negative bacteria respectively. Experimental conditions were as detailed above. Approximately 80% of the water in each replicate was renewed at test initiation and on the second exposure day. At test end, a subset of fish were used for swimming assessments and remaining fish snap-frozen and stored at –80°C for subsequent biomarker analyses.

Swimming assessments were performed at test takedown. Fish were placed in rectangular tanks (12 x 6 x 9 cm) containing control water, and allowed to acclimate for 5 min. Three minute video imaging, recorded in MPEG-2 format, was performed at 30 frames per second using a black and white Panasonic CCTV camera (12VDC) connected to a laptop computer via a USB framegrabber (Model WinTV-HVR 950). Video analysis was carried out using Ethovision XT (Version 6.1.326, Noldus Information Technology). Average velocity was determined for each fish by analyzing a total of 72 seconds per video test. One-way ANOVA was used to assess differences in gene expression and Dunnett's multiple comparison post hoc tests were used to compare exposed treatments to controls.

Experimental physicochemistry:

For all exposure tests, water temperature, pH, and DO were measured daily. Ammonia nitrogen (NH_4^+-N) concentrations were measured prior to each water renewal and at test termination. Dissolved copper analyses were carried out by the California Department of Fish and Game – Water Pollution Laboratory, Rancho Cordova, CA.

Microarray application

Development of the delta smelt microarray was described in Connon *et al.* (2009) [15], briefly we have constructed a cDNA microarray with 8,448 expressed sequence tags (ESTs) which were pin-printed in duplicate onto epoxysilane coated glass slides.

RNA isolation, cDNA synthesis and fluorescence labeling.

RNA was extracted from whole, individual organisms, using Trizol Reagent (Invitrogen) as per manufacturer's guidelines. Fifteen micrograms of total RNA were used for cDNA synthesis, spiked with control RNA (CAB, RCA, RBCL and LTP4 (SpotReport, Stratagene) and labeled with Alexa fluor dyes, using SuperScript^{tm} Plus Indirect cDNA labeling System (Invitrogen). Microarray assessments were carried out using three replicate treatments. Each experimental sample or control was combined with a reference pool cDNA prior to hybridization using an automated Tecan HS4800 hybridization station. Slides were scanned using a GenePix 4000B scanner (Axon Instruments).

Microarray images and data from exposed delta smelt can be accessed at http://www.vetmed.ucdavis.edu/apc/WernerLab/subpage/pelagic_organism_decline.html; POD archive data.

Microarray Analyses

Normalization and analytical methods are described in [15, 33]. In brief, 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 α-outlier-generating model was used to identify differentially expressed genes by applying the following decision rule for multiple-slide data analysis: a given gene was selected as a candidate if it was consistently detected as up- or downregulated in 4 of 4 replicates (raw p-value = 0.0625 using exact binomial test and considering outcomes as Bernoulli trials). The approach did not use scale estimation for statistical inference and, therefore did not require between-slide normalization. We used a higher than usual cut-off point of 0.0625, due to normal microarray normalization stringencies, as the purpose of this investigation was to identify genes that could be assessed as probable qPCR based molecular biomarkers for future monitoring programs (see below).

Sequencing and Annotation

Sequencing was carried out at the CA&ES Genomic Facility, UC Davis. Basic Local Alignment Search Tool; translated nucleotide (BLASTx) searches were performed on specific fragments that responded significantly to the exposure treatments. Only genes that were differentially expressed following exposure were sequenced. Sequences were annotated according to homologies to protein database searches using translated nucleotide sequences and direct nucleotide queries (http://blast.ncbi.nlm.nih.gov/Blast.cgi). Sequences were only annotated if they were found to have a BLASTx match with the expect value smaller than $1x10^{-5}$ and a score above 50.

Functional Classifications

Differentially expressed genes were classified according to the Kyoto Encyclopedia of Genes and genomes (KEGG - http://www.genome.jp/kegg/kegg2.html) and Gene Ontology (GO - http://www.uniprot.org/uniprot), and information gathered from literature, into functional groups. Classification was carried out based on gene expression changes in respect of control subjects, regardless of whether these were up- or downregulated. Specific genes of interest were selected for further investigation using quantitative PCR (see below).

Biomarker development

Genes were selected according to level of expression significance, knowledge base from literature, and functional classification. Primer and probes for qPCR analyses were designed using Roche Universal Probe Library Assay Design Center (https://www.roche-applied-science.com). Designed primers were obtained from Eurofins MWG Operon (http://www.eurofinsdna.com), and TaqMan probes were supplied by Roche. Sequences for all genes assessed by qPCR analyses have been submitted to GenBank (http://www.ncbi.nlm.nih.gov). Primers and probes for investigated biomarkers are detailed in table 1.

Quantitative PCR

Complementary cDNA was synthesized using 1.0 µg total RNA, with random primers and SuperScript® III reverse transcriptase (Invitrogen), and diluted to a total of 120 µl with nuclease free water to generate sufficient template for qPCR analysis. TaqMan Universal PCR Mastermix (Applied Biosystems) was used in q-PCR amplifications in a reaction containing 10mMTris–HCl (pH 8.3), 50mM KCl, 5mM MgCl2, 2.5mM deoxynucleotide triphosphates, 0.625U AmpliTaq Gold DNA polymerase per reaction, 0.25U AmpErase UNG per reaction and 5µL of cDNA sample in a final volume of 12µL. 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 60s 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, CT). SDS 2.2.1 software (Applied Biosystems) was used to quantify transcription.

Statistical analyses

We used the geNorm algorithm [34] to estimate the variability of the reference genes, and to determine an optimal normalization gene.

Quantitative PCR data was analyzed using the relative quantification 2(-Delta Delta CT) method [35]. Expression was calculated relative to a-actin determined by GeNorm as the least variable gene in this study. Quantitative PCR data were not normally distributed, therefore, significant differences in gene expression, relative to the unexposed controls, were assessed using two-tailed Mann-Whitney U test, single comparison alpha = 0.05, with

Bonferroni's correction experiment-wide alpha = 0.15, treating each gene as a separate experiment.

Results

Fish Exposures and swimming assessments

i. Exposures used for microarray analyses:

Juvenile delta smelt (90-d old) were highly sensitive to copper exposure (table 2), resulting in an estimated $LC_{50\text{-}7d}$= 17.8 $\mu g.Cu^{2+}.L^{-1}$. Surviving fish from the 42.0 $\mu g.Cu^{2+}.L^{-1}$ exposure treatment, were pooled, per replicate, and used for microarray analysis, against controls, in order to obtain copper specific genomic expression information (see microarray responses, below).

ii. Exposures used for quantitative PCR and swimming video-analyses:

Exposure of 47-d old larval delta smelt to copper for 4 days resulted in an $LC_{50\text{-}96h}$ = 80.4 $\mu g.Cu^{2+}.L^{-1}$ (table 2). Differences in copper sensitivity between juvenile and larval exposures were attributed not only to age and size, but also to temperature and conductivity, which are known to affect metal uptake and toxicity [36, 37]. Due to high mortality resulting at the highest copper concentration, surviving fish numbers were not sufficient for use in qPCR tests and thus were discarded from further analysis.

Video-analysis of larval swimming performance (figure 1) has demonstrated an overall effect of copper exposure resulting in declining velocity with increasing copper concentration (r=-0.911).

Experimental water physicochemistry:

Water physicochemical parameters for all tests remained stable throughout the exposures (table 3) except for a lower ammonium-nitrogen concentration in the 42.0 $\mu g.CuCl_2.L^{-1}$ 7 day test used for microarray analysis, which was attributed to a lower number of remaining fish due to high mortality. Discrepancy in total ammonium (NH_4^+N) between exposure sets are attributed to development stage, age and size differences between juvenile and larval delta smelt.

Comparative toxicological data:

Differences in the above reported toxicity between juvenile and larval delta smelt (table 2) are due to exposure duration discrepancies and developmental stage, but variations in experimental conditions such as differences in water conductivity, pH, and temperature, also contributed to the higher copper toxicity to juveniles. Furthermore, the larval exposure tests were carried out using anitibiotic treatments. Though juvenile delta smelt appear more sensitive to copper exposure than do larvae, acute toxicity results are thus inconclusive, however, genomic responses have been analyzed successfully in a comparative manner (see below).

Microarray responses

Differentially expressed genes resulting for exposure to 42.0 μg.Cu^{2+}.L^{-1} are presented in table 4. A functional classification based on KEGG and GO of up- and down-regulated genes responding to copper exposure are presented in figure 2.

Copper exposure primarily impacted on neuromuscular activity, affecting muscle integrity and contraction activity (e.g. creatine kinase, myozenin, sarcoendoplasmic reticulum calcium ATPase, titin a), neurological effects resulting in calcium and phosphate signaling (e.g. m-calpain, cyclophilin-a) and nerve maintenance (hemopexin and aspartoacylase). Copper is reported to inhibit iron storage through interaction with peroxidases causing oxidative stress which leads to disruption in calcium homeostasis [38]; resulting from effects upon Na^{+}/Ca^{2+} membrane exchange, thus elevating intracellular Ca^{2+} [39]. Digestion was also affected by copper exposure, including genes encoding a number of proteins involved in glycolysis, cholesterol efflux, lipid transport, chymotrypsin activity, proteolysis (e.g. amylase-3, gastric chitinase). Other responses indicate compromised immunity (e.g. TNF, TGF-β), and cellular homeostasis and tumor malignancy (e.g. vitronectin), changes in expression of these proteins have been implicated in a variety of diseases.

Gene classification from KEGG Orthology analyses also indicate effects on expression of genes encoding proteins involved in the Peroxisome Proliferator-Activated Receptor (PPAR) pathway, receptors that function as transcription factors regulating gene expression, playing an essential role in the regulation of cellular differentiation, development, metabolism of carbohydrate, lipids and proteins, and tumorgenesis. This pathway integrates the majority of genes classified into digestion and metabolism.

Molecular Biomarker responses

Genes were selected according to level of expression significance and to represent the identified functional classifications. Thus genes involved in muscular, neurological, digestive and immune system functional groups were further investigated, using q PCR, and assessed as probable biomarkers of copper exposure in *H. transpacificus*.

Results confirm microarray identified functional responses. Quantitative PCR verified copper elicited responses in neuromuscular, digestive and immune system functions (figure 3a-d), with significant differences in expression of muscle α-actin, ASPA, hemopexin, chitinase and TNF ($p<0.05$). Remaining assessed genes displayed dose response relationships, and/or differences in expression trends, but were not statistically significant compared to controls. Though not statistically significant in their expression level, responses from these genes directly facilitate the interpretation of functionally affected systems and are thus interpreted with biological significance.

Muscular structure and activity:

Muscular structure and integrity appears to have been affected by copper exposure (figure 3a), as indicated by effects on contractile muscle systems; α-actin and myozenin. Skeletal

α-actin, significantly downregulated at all concentrations, is also reported to induce expression of a number of other myogenic genes essential for muscle formation [40]. Thus actin may serve as wholistic muscle integrity and functioning effect biomarker. Myozenin is a Z-line, α-actinin- and γ-filamin-binding protein expressed predominantly in skeletal muscle, and has been suggested as a biomarker for muscular dystrophy and other neuromuscular disorders [41].

Muscle activity was also affected by copper exposure, altering Ca^{2+} homeostasis, denoted by SER Ca ATPase and Creatine Kinase altered expression. SER Ca ATPase a muscle sarcoendoplasmic reticulum (SR) calcium ATPase pump, responsible for the transfer of calcium from the cytoplasm into the SR after muscle activity [42, 43], was downregulated by copper exposure in a dose dependent manner. This is suggestive of a potential biomarker of muscular activity, indicative of mobility impairments, and likely apoptotic responses. Downregulation of SER Ca ATPase mRNA signifies a decrease in enzyme synthesis, likely causing a decrease in Ca^{2+} in the SR lumen. Disruption of Ca^{2+} homeostasis within the SR has been postulated as an early warning sign of apoptosis, thus inhibition of SER Ca ATPase could lead to cell death [44, 45].

A further biomarker assessed to measure muscular effects of copper exposure was Creatine kinase (CK). Protein concentrations are used as a diagnosis of diseases like cardiac infarction and skeletal muscle necrosis [46]. It is specifically bound to sarcoendoplasmic reticulum, and regulates calcium uptake and ATP/ADP ratios [47], thus is directly linked with SER Ca ATPase and involved in muscle activity. Though not statistically significant, CK expression was also reduced by increasing copper concentration, in a similar manner displayed by SER Ca and α-actin, suggesting a decline in calcium regulation and overall energetic activity.

Neurological activity:

Copper exposure is known to affect the nervous system through the formation of reactive radicals [38, 48], an effect that was sustained in this study (figure 3b), through expression of hemopexin, which was significantly upregulated at lower concentrations. Hemopexin is synthesized by Schwann cells following nerve injury [49], has been reported in the peripheral nervous system, and is specifically regulated during repair [50]. The measured downregulation at higher concentrations could imply inhibition of repair mechanisms.

Wallerian degeneration generally occurs following axonal injury and is critical for its repair. This is characterized by axonal and myelin degeneration [51], thus Aspartoacylase (ASPA), a gene identified in a previous study [15], was investigated as it is expressed in myelin sheaths and involved in their maintenance. Because of its functional proximity to hemopexin, it was chosen to further assess neurological damage, however, ASPA did not respond significantly to copper exposure, but displayed an increase at lower concentrations, supporting measured differences in hemopexin transcription. Both hemopexin and ASPA expression were elevated at lower concentrations and reduced at higher concentrations. Effects of copper exposure on calcium availability were further corroborated by changes in m-Calpain expression (figure 3b). m-Calpain is a calcium-dependent cysteine protease, known to co-localize with a calcium-sensing receptor, where calcium not only activates the

m-Calpain enzyme, but also causes it to undergo autolysis through subunit dissociation [52]. The physiological roles of calpains are still poorly understood, however, they have been shown to control cell fusions in myoblasts, playing an important role during myogenesis and thus muscle regeneration [53]. Interestingly, activation of m-Calpain in the peripheral nervous system has been also reported to be involved in Wallerian degeneration, with increased expression being initiated following nerve injury [54], which was also indicated by changes in ASPA expression levels (see above). Thus m-Calpain is a potential biomarker of neuromuscular activity, and as such clustered into both muscular and neurological responses. However due to high variability in control subjects, and the low number of replicates available in this study, changes in expression were not significantly different in this test.

Digestion:

Heavy metals are known to affect digestion [55]. Copper exposure resulted in significant responses in transcription of genes involved in delta smelt digestion (figure 3c). α-Amylase is a digestive enzyme that hydrolyses starch into maltose [56]. It was downregulated, significantly correlating (r=0.978) with increasing copper concentration. Downregulation of α-amylase transcription has previously been associated with copper exposure [57-59]. Chitinase, an enzyme required in the digestion of chitin structures in the exoskeleton of crustacean and many insects, was significantly upregulated on exposure to copper. It has recently been found that fish express chitinase in their guts [60], thus hypothesized to be involved in arthropod digestion. Chitin is also a component of some bacterial cell walls [61], and chitinase has been reported as a hydrolase involved in the defense against Gram positive bacteria and fungal pathogens, particularly during larval stages [60], though this role has not fully been established [62].

Immune responses:

Microarray analysis also identified effects upon the immune system that were confirmed through qPCR (figure 3d). Tumor Necrosis Factor, a proinflammatory cytokine, was significantly downregulated by copper exposure, indicating a compromised immune system. Produced in many cell types, TNF plays an important role in immunity and inflammation, and in the control of cell proliferation, differentiation and apoptosis [63] and its downregulation has been implicated with various diseases such as Crohn's, arthritis, multiple-sclerosis and Alzheimer's [63, 64] . Balkwill et al [64], indicate that TNF knock-out causes autoreactive T Cells regulation, resulting epitope spreading, thus leading to a state of disease. Transforming growth factor-β is an anti-inflammatory cytokine expressed functionally during development and in tissue maintenance and homeostasis regulating proliferation and differentiation, cell survival and apoptosis [65, 66]. Upregulation of TGF-b has been linked with neurodegenerative diseases and ischemic injuries [67] and interestingly is reported to induce muscle α-actin expression [68], which was downregulated by copper exposure. TGF-β displayed an upregulation at lower levels of copper exposure, in similar trends observed in ASPA and hemopexin, further suggesting immune responses resulting from probable neurological or signaling impairments.

Discussion

Molecular biomarkers in monitoring programs:

Molecular biomarkers have, for many years, successfully been used in human medicine as diagnostic tools, for example the assessment of prostate specific antigen (PSA) gene expression as an indicator of prostate cancer [69], and are increasingly being used in the pharmaceutical arena to assess the mode of action, safety, efficiency and targeted effects of novel and developed drugs [70]. Research in environmental toxicology has been concentrating on attributing or defining an ecological relevance to responses measured using biomarkers, through measuring responses at the individual level and attempting to extrapolate results to population responses. This work has been the subject of extensive critical discussion that has successfully strengthened biomarker research, but has concurrently discouraged wide spread application in field studies. However, a limited number of researchers have successfully applied molecular biomarkers in field studies, identifying contaminant stress induced responses [71, 72] and metal contamination effects [73] in various field collected aquatic organisms. Others report successful applications on field deployed organisms [74, 75]

In most aquatic organisms, and particularly in fish, a number of life cycle characteristics are dependent on swimming behavior, from respiration to reproduction, thus swimming performance in fish, is likely the single most significant environmentally relevant parameter that can be measured, as it is indicative of effects upon behavior. Thus, effects upon individual fish, however minimal, will reduce reproductive success of a population. It is clear that neuromuscular alterations will have significant effects on swimming performance. In this study we confirm that a suite of molecular biomarkers, designed specifically to address neuromuscular disturbances, can successfully indicate, allow interpretation of, and correlate with effects on swimming ability, but are also informative of the mode of action of stressors upon impinging on this ability.

The use of biomarkers to specifically address key health parameters have extensively been investigated, through proteomics, a global genomics approach, or with the application of molecular probes, such as neuromuscular activity, digestion and immune responses, as in this study, or with the addition of developmental assessments, such as links with endocrine responses, growth and sexual development. In the search for biomarkers of effect and exposure, it is traditional to ascertain as useful, only those whose functional responses correlate with exposure concentration. However, hormetic or biphasic dose responses appear to be indicative of changes in homeostasis [76, 77], and as such should be treated as an invaluable technique to identify concentrations at which organisms can no longer compensate adequately to exposure. It is our contention that biphasic responses can therefore be more informative that dose responsive biomarkers, which are solely indicative of exposures, without identifying lowest concentrations at which detrimental effects may occur. Thus a suite of biomarkers, both biphasic and dose respondent should be utilized in conjunction in order to elucidate effects upon an organism. We have previously demonstrated the ability to link neurological effects from pyrethroid exposures in the delta smelt, with changes in gene expression that correlated with swimming behavior [15]. In order to attribute environmental relevance to molecular biomarkers, research should concentrate on their application in field studies with the aim of developing monitoring

programs. Only through the inclusion of these techniques in monitoring programs, can biomarkers truly be evaluated.

Molecular biomarkers in the delta smelt:

The delta smelt are highly sensitive to handling and extremely difficult to work with under laboratory conditions, and these studies are not necessarily informative of what the organism are exposed to in their habitat. Thus reliable biomarkers for determining the health status and exposure history of delta smelt, in field based studies are essential. Molecular biomarkers such as those identified in this study, could therefore be coupled to, and carried out in collaboration with, the Californian Department of Fish and Game (DFG) annual townet surveys and monitoring programs (http://www.dfg.ca.gov/delta). Thus creating an informative database of genomic responses indicative of delta smelt health status, aiding the process of toxicity identification and evaluation through the identification of contaminant specific responses within complex chemical mixtures. Base line gene expression, for comparative purposes, could be generated from assessing hatchery raised larval, juvenile and adult delta smelt, alongside temporal, site specific variations in townet surveyed wild fish.

Biomarker suite recommendations for field based monitoring studies in the delta smelt:

A number of delta smelt genes, identified in this and previous studies [15] have been demonstrated as specific and informative biomarkers to warrant their application in field studies. Utilizing the molecular biomarkers identified this far, we are able to identify and further understand neuromuscular effects resulting from copper and pesticide exposures. Combining gene expression changes in aspartoacylase and hemopexin, for example, has proven successful in the identification of neurological insults, suggesting degradation of axon myelin sheaths and nerve repair [15]. Biphasic changes in expression seemingly differentiate homeostatic responses, allowing repair mechanisms, from concentrations which are chronically detrimental, as implied by aspartoacylase and hemopexin responses in this study. Effects of copper on muscular activity and integrity can also be ascertained through the resulting downregulation of SER Ca ATPase and α-actin respectively. These are genomic measurements that statistically correlate with declining swimming behavior and are biologically significant in the interpretation of exposure effects of neurotoxicants. Furthermore, the health status of an organism can be assessed in terms of digestion capacity and immune system functioning. Additional to the qPCR based biomarkers already described, a number of prospective genes have been identified with the microarray application, and will be isolated from future studies, to expand genomic information towards a suite of functionally classified biomarkers to be in monitoring programs and identify classes of contaminants present in the delta smelt' habitat range that may be responsible for toxicity. Thus, the presently assessed suite of biomarkers could be applied to field studies, comprising site specific collected water sample exposures within a laboratory conditions, as well as upon wild specimens caught during townet surveys.

Linking molecular responses to copper exposure with condition indicators:

The short-term copper exposure (4-d) in this study resulted in an overall decrease in swimming velocity in larval delta smelt with increasing concentration. The reduction in swimming activity can be explained by alterations measured by all neuromuscular molecular biomarkers, as these indicate the mechanisms of action of copper upon the delta smelt. SER Ca ATPase was a particularly informative gene, as the measured downregulation confirmed interference of copper with Ca^{2+} homeostasis, neurological signaling and muscle activity. The strong downregulation of α-Actin, at all concentrations, sustains reported effects of copper on contractile muscle proteins [59], further supported by increases in m-Calpain expression, involved in muscle regeneration [53].

Food consumption was not measured in this test, thus it is not clear from this study whether the measured digestive changes are directly due to copper exposure acting upon specific enzymes, or whether this is a result of altered swimming performance impinging on prey capture and ingestion. Under unexposed conditions, elevated levels of chitinase would likely signify higher levels of ingestion. It is unlikely that copper exposed fish had a higher consumption level, thus our contention is that copper directly acts upon digestion either impeding the proteolytic process through synthesis inhibition, enzyme degradation or mRNA expression inhibition, as indicated through amylase expression.

Further responses to copper exposure indicate compromised immune system, with specific links to the central nervous system, as suggested by the measured upregulation of TGF-β, reported to be linked to neurodegenerative diseases [67], and its involvement in the induction of contractile muscle protein [68] further supports neuromuscular damage.

Copper concentrations used in this study, though high, are not uncommon [58], especially the lowest concentration investigated. Results from this study are indicative of short-term exposure responses. Bioaccumulation properties of heavy metals are well researched [78], and we extrapolate that longer-term exposures to lower levels of copper are likely to have similar effects on swimming performance and altering the overall chances of delta smelt survival in the wild. Indisputably, the primary reason for the decline in number of pelagic organism in the Sacramento-San Joaquin Delta is directly related to water exports [22-24], however, organisms that manage to survive this habitat destruction, are exposed to elevated concentrations of contaminant resulting from industrial, agricultural and urban pollution, lower water flows combined with a lesser dilution rate. Management systems to monitor the extent of change resulting from anthropogenic loads are essential, and this study enhances the argument for the use of a suite of molecular biomarkers as a successful approach towards identifying effects and causal factors of species decline.

Tables and Figures:

Table 1. Molecular biomarkers: Primer and probe sequences used for quantitative-PCR analyses of gene expression in *H. transpacificus*. * indicates reference gene.

Table 2. Physicochemical parameters of control and test waters from 4 and 7-d copper exposures.

Table 3. Acute toxicity data from copper exposed 90-d old juvenile and 47 d-old larval delta smelt.

Table 4. Annotation, gene ontology and regulation of gene expression, in juvenile delta smelt exposed to copper (42.0 $\mu g.Cu^{2+}.L^{-1}$).

Figure 1. Swimming performance of 47-d old larval delta smelt exposed to copper for 96 h.

Figure 2. Microarray responses**:** systematic analysis of KEGG Orthology and Gene Ontology based functional classification of delta smelt genes significantly differing in juveniles exposed to copper (42.0 $\mu g.Cu^{2+}.L^{-1}$) for 7-days.

Figure 3. Quantitative PCR expression assessments of selected delta smelt genes responding to copper exposed larvae. * indicates tests significant differences ($p<0.05$ at. n= 9, 9, 8, and 5, for concentrations 1.6, 24.0, 52.4 and 105.0 respectively).

Gene	GenBank Accession No.	q-PCR Primer Sequences	Roche Probe No.
Myozenin	FJ711583	F CCAATGTCGTGCTGGTACACC R CTGCCAGACATTGATGTAGCCA	106
Creatine Kinase	FJ711584	F CGATCGGCGTTGGAGATG R GCCAAGTTCAACGAGATTCTGG	163
SER-Ca ATPase	Submitted	F CATGATCATTGGGGGAGCA R TGCTGTGATGACAACGAGGAC	148
α-Actin	Submitted	F CCTGCCTCGTCGTACTCCTG R CATCCTGGCTTCCCTGTCC	11
m-Calpain	Submitted	F CCCTCCGACATGGGAAGAGT R ACCAACTATGCCTTGCCCAA	30
Aspartoacylase	FJ711577	F GGAGGCACACATGGGAATG R CTTCCTCTGAATCTCTGTTCCATTATC	109
Hemopexin	FJ711579	F CATGCACTACGAGGACGACAAG R TGGTAGTAGCTGAACACCTTGCTG	143
Chitinase	Submitted	F TGTGATCAAGTTCCTCCGTCAGT R CCGGGGTATTCCCAGTCAAT	147
α-Amylase	Submitted	F GATCACCATGTTCTTGATCTGACG R CCATCAATCCTGACCAAACCTG	99
TGF-β	Submitted	F CAACGGCATAGTGCATGTGG R GAATGTGTGCACGTTGTTGGT	76
TNF-decoy receptor	Submitted	F CTTTTTCCGCTGTTCCATGTTC R GTTACCAGCATACGCAGTGTCC	2
β-Actin*	Submitted	F TGCCACAGGACTCCATACC R CATCGGCAACGAGAGGTT	12

Table 1.

Treatment	Temp (°C)	pH	EC ($\mu S.cm^{-1}$)	DO ($mg.L^{-1}$)	NH_4^+-N ($mg.L^{-1}$)	Weight (g)	Length (cm)
i. Exposures used for microarray analysis (90-d old juvenile, 7-d exposure):							
Control water	21	8.40	431	8.8	0.28	0.24	3.42
4.2 $\mu g.Cu^{2+}.L^{-1}$	21	8.49	456	8.7	0.24	0.24	3.53
8.4 $\mu g.Cu^{2+}.L^{-1}$	21	8.48	461	9.0	0.23	0.23	3.49
21.0 $\mu g.Cu^{2+}.L^{-1}$	21	8.46	455	8.8	0.37	0.26	3.57
42.0 $\mu g.Cu^{2+}.L^{-1}$	21	8.39	457	8.9	0.14	0.26	3.52
Experimental Mean	21	8.44	452	8.84	0.25	0.25	3.51
ii. Exposures used for quantitative PCR and swimming ability analyses (47-d old larvae, 4-d exposure):							
Control water	17	7.86	931	9.1	0.025	N/A	N/A
24.0 $\mu g.Cu^{2+}.L^{-1}$	17	7.84	926	9.3	0.037	N/A	N/A
52.4 $\mu g.Cu^{2+}.L^{-1}$	17	7.89	927	9.4	0.057	N/A	N/A
105.0 $\mu g.Cu^{2+}.L^{-1}$	17	7.93	931	9.4	0.047	N/A	N/A
213.0 $\mu g.Cu^{2+}.L^{-1}$	17	7.89	931	9.5	0.033	N/A	N/A
Experimental Mean	17	7.88	929	9.34	0.039	N/A	N/A

Table 2.

Endpoint	**Juvenile exposure**				**Larval exposure**	
	96-h	*95% CI*	*7-d*	*95% CI*	*96-h*	*95% CI*
NOEC	8.4	-	8.4	-	53.0	-
LOEC	21.0	-	21.0	-	106.0	-
LC10	9.6	4.2 - 11.4	9.0	4.2 - 10.3	9.3	27.0 - 77.8
LC25	13.4	10.6 - 18.0	11.7	9.7 - 13.0	44.8	27.0 - 83.1
LC50	25.2	16.4 - 35.4	17.8	14.4 - 22.4	80.4	48.7 - 227.2

Table 3.

Gene most similar to	Species Match	Accession No	E-Value	Score	GO	Fold-change
pancreatic protein with two somatomedin B domains	*Paralichthys olivaceus*	BAA88246	2.00E-95	352	GO:0005179	7.54
cell division cycle 14 homolog A	*Danio rerio*	CAP09233	3.00E-19	99	GO:0004725	5.76
corticotropin-lipotropin A precursor	*Oncorhynchus mykiss*	Q04617	7.00E-63	244	GO:0005179	5.20
elastase 2-like protein	*Sparus aurata*	AAT45251	2.00E-89	332	GO:0006508	5.08
actin alpha 2, skeletal muscle	*Pagrus major*	BAF80060	1.00E-94	384	GO:0003774	4.88
phosphoglucose isomerase-2	*Plecoglossus altivelis*	BAF91566	1.00E-120	435	GO:0006096	4.86
apolipoprotein A-I-2 precursor	*Oncorhynchus mykiss*	O57524	4.00E-71	271	GO:0033344	4.81
pepsinogen A form IIa	*Pseudopleuronectes americanus*	AAD56283	1.00E-105	384	GO:0004194	4.65
arachidonate 12-lipoxygenase	*Danio rerio*	NP_955912	4.00E-23	112	GO:0004052	4.42
chitinase1	*Paralichthys olivaceus*	BAD15059	1.00E-127	458	GO:0004568	4.25
lipoxygenase 12R (PREDICTED: similar to)	*Ornithorhynchus anatinus*	XP_001518171	8.00E-06	55	GO:0016165	4.17
apolipoprotein Eb	*Danio rerio*	NP_571173	2.00E-38	162	GO:0033344	4.16
SPARC: secreted protein, acidic, rich in cysteine	*Danio rerio*	AAT01213	2.00E-31	139	GO:0006816	4.14
pepsin A2	*Trematomus bernacchii*	CAD80096	2.00E-88	253	GO:0004194	4.05
apolipoprotein A-I-1 precursor (Apo-AI-1)	*Oncorhynchus mykiss*	O57523	8.00E-76	286	GO:0033344	3.99
chymotrypsinogen 2-like protein	*Sparus aurata*	AAT45254	1.00E-20	101	GO:0004263	3.93
myozenin 1	*Danio rerio*	NP_991241	2.00E-25	119	GO:0030346	3.91
NADH dehydrogenase subunit 5	*Osmerus mordax*	ABI35911	1.00E-107	390	GO:0008137	3.88
astacin like metallo-protease	*Oryzias latipes*	NP_001098207	2.00E-83	311	GO:0008533	3.87
Hect domain and RLD 4 (PREDICTED: similar to)	*Danio rerio*	XP_685685	7.00E-76	286	GO:0006512	3.76
actin, alpha 2, smooth muscle, aorta	*Danio rerio*	AAH75896	1.00E-107	391	GO:0003774	3.75
chitinase	*Oncorhynchus mykiss*	CAD59687	9.00E-68	260	GO:0004568	3.75
F-type lectin	*Morone saxatilis*	ABB29997	1.00E-46	188	GO:0016467	3.73
Pgk1(phosphoglycerate kinase 1) protein	*Danio rerio*	AAH65888	9.00E-84	313	GO:0006096	3.67
aldolase a, fructose-bisphosphate	*Danio rerio*	NP_919358	1.00E-124	447	GO:0006096	3.47
NADH dehydrogenase subunit 5	*Osmerus mordax*	ABI35911	5.00E-94	308	GO:0008137	3.47
pepsinogen C (progastricsin)	*Salvelinus fontinalis*	AAG35646	1.00E-107	390	GO:0004194	3.41
amylase-3 protein	*Tetraodon nigroviridis*	CAC87127	3.00E-54	213	GO:0004556	3.36
simple type II keratin K8b (S2)	*Oncorhynchus mykiss*	CAA63300	3.00E-74	281	GO:0005882	3.28
glutamate dehydrogenase 1	*Danio rerio*	NP_955839	1.00E-107	392	GO:0004352	3.24
α-amylase	*Pseudopleuronectes americanus*	AAF65827	1.00E-144	513	GO:0004556	3.06
pepsinogen	*Paralichthys olivaceus*	BAC87742	3.00E-77	291	GO:0004194	3.04
NADH dehydrogenase subunit 6	*Salangichthys microdon*	NP_795843	1.00E-107	392	GO:0008137	3.03
Hemopexin	*Danio rerio*	NP_001104617	1.00E-59	233	GO:0046872	3.02
gamma2-synuclein	*Takifugu rubripes*	NP_001029019	2.00E-41	172	GO:0030424	2.93
actin, alpha, cardiac muscle 1 like	*Danio rerio*	NP_001001409	1.00E-141	503	GO:0003774	2.92
cardiac muscle ATP synthase,alpha 1,	*Danio rerio*	NP_001070823	7.00E-62	240	GO:0015662	2.91
selenoprotein P, 1a	*Danio rerio*	NP_840082	1.00E-53	213	GO:0001887	2.86
intestinal fatty acid binding protein	*Danio rerio*	AAF00925	3.00E-56	221	GO:0008289	2.82
L-arginine:glycine amidinotransferase	*Danio rerio*	NP_955825	5.00E-83	310	GO:0016740	2.76
apolipoprotein A-IV	*Danio rerio*	AAH93239	1.00E-73	279	GO:0006869	2.72
peptidylprolyl isomerase A (cyclophilin)	*Danio rerio*	AAQ91263	2.00E-74	282	GO:0003755	2.66
histone methyltransferase SmyD1b	*Danio rerio*	ABC54714	1.00E-108	394	GO:0030239	2.62
sarcoendoplasmic reticulum calcium ATPase	*Silurus lanzhouensis*	ABG90496	8.00E-79	297	GO:0006937	2.47
1-acylglycerol-3-phosphate O-acyltransferase 3	*Danio rerio*	NP_998590	4.00E-68	261	GO:0003841	2.36
chitin binding Peritrophin-A domain	*Danio rerio*	AAH45331	4.00E-69	264	GO:0016490	2.34
apolipoprotein A-I	*Danio rerio*	NP_571203	1.00E-81	306	GO:0033344	2.28
calpain 1 protein	*Danio rerio*	AAH91999	2.00E-68	262	GO:0005509	2.27
apolipoprotein B	*Salmo salar*	CAA57449	3.00E-24	115	GO:0030301	2.23
sarcoendoplasmic reticulum calcium ATPase	*Makaira nigricans*	AAB08097	1.00E-83	313	GO:0006937	2.22
muscle creatine kinase	*Danio rerio*	CAM16434	1.00E-112	406	GO:0004111	2.21
transmembrane protein 38A	*Danio rerio*	NP_957194	8.00E-81	303	GO:0005267	2.21
tripartite motif-containing 45	*Xenopus tropicalis*	NP_001011026	3.00E-27	125	GO:0046872	2.20
titin a	*Danio rerio*	ABG48500	3.00E-88	328	GO:0031432	2.19
c1q-like protein	*Dissostichus mawsoni*	ABN45966	3.00E-38	162	GO:0006817	2.17
apolipoprotein CII	*Oncorhynchus mykiss*	AAG11410	1.00E-19	100	GO:0006869	2.02
guanine nucleotide binding protein (G protein), beta 1	*Danio rerio*	NP_997774	1.00E-117	424	GO:0003924	1.88
alpha tubulin, (protein LOC573122)	*Danio rerio*	NP_001098596	1.00E-120	434	GO:0007018	1.86
DAZAP2-like protein (deleted in azoospermia-associated)	*Takifugu rubripes*	NP_001072102	5.00E-59	230	GO:0030154	1.86

acyl-CoA synthetase long-chain family member 5	*Tetraodon nigroviridis*	CAG06540	1.00E-102	375	GO:0004467	1.83
carboxypeptidase H	*Paralichthys olivaceus*	AAO92752	1.00E-82	309	GO:0004183	1.82
apolipoprotein	*Tetraodon nigroviridis*	CAG03661	1.00E-38	78	GO:0030301	1.80
transforming growth factor, beta-induced	*Danio rerio*	NP_878282	3.00E-21	105	GO:0008083	1.59
neurotransmitter transporter, glycine, member 9 (SLC6A9)	*Danio rerio*	CAM14205	1.00E-100	367	GO:0006836	0.65
calcium binding protein 39	*Danio rerio*	NP_998666	1.00E-76	290	GO:0019855	0.63
cytochrome P450, family 46, subfamily A, polypeptide 1	*Danio rerio*	NP_001018358	2.00E-65	252	GO:0004497	0.63
E3 ubiquitin-protein ligase MARCH2	*Danio rerio*	Q1LVZ2	2.00E-87	325	GO:0006512	0.60
calcitonin receptor-like receptor	*Oncorhynchus gorbuscha*	CAD48406	5.00E-56	221	GO:0004948	0.59
dopachrome tautomerase	*Salmo salar*	ABD73808	1.00E-85	318	GO:0016491	0.56
tetraspanin 7b	*Danio rerio*	NP_001005581	1.00E-110	400	GO:0022857	0.54
isocitrate dehydrogenase 3 (NAD+) gamma	*Danio rerio*	NP_001017713	2.00E-14	83	GO:0016616	0.53
cofilin 2 (muscle)	*Danio rerio*	NP_991263	5.00E-84	314	GO:0003779	0.50
m-calpain	*Oncorhynchus mykiss*	BAD77825	1.00E-108	396	GO:0005509	0.50
zona pellucida protein X	*Sparus aurata*	AAY21008	1.00E-68	263	GO:0032190	0.50
suppressor of ypt1	*Danrio rerio*	NP_878281	1.00E-122	442	GO:0016192	0.47
thioredoxin-like 1	*Danio rerio*	NP_957432	1.00E-107	391	GO:0045454	0.44
lactase-phlorizin hydrolase (PREDICTED: similar to)	*Danio rerio*	XP_001336765	1.00E-110	401	GO:0005975	0.43
potassium channel tetramerisation domain containing 5	*Danio rerio*	NP_996932	2.00E-76	288	GO:0005249	0.39
zinc finger protein 503	*Danio rerio*	NP_942137	3.00E-63	245	GO:0003676	0.39
ornithine decarboxylase	*Paralichthys olivaceus*	AAO92750	9.00E-67	256	GO:0006596	0.35
proteasome subunit alpha type 7	*Danio rerio*	NP_998331	1.00E-112	409	GO:0030163	0.35
proteasome (prosome, macropain) 26S subunit, ATPase, 4	*Danio rerio*	AAI53480	1.00E-109	396	GO:0030163	0.34
TNF (tumor necrosis factor) decoy receptor	*Oncorhynchus mykiss*	AAK91758	5.00E-67	257	GO:0004872	0.26
APEX nuclease (apurinic/apyrimidinic endonuclease) 2	*Xenopus tropicalis*	NP_001006804	6.00E-25	118	GO:0006281	0.22

Table 4.

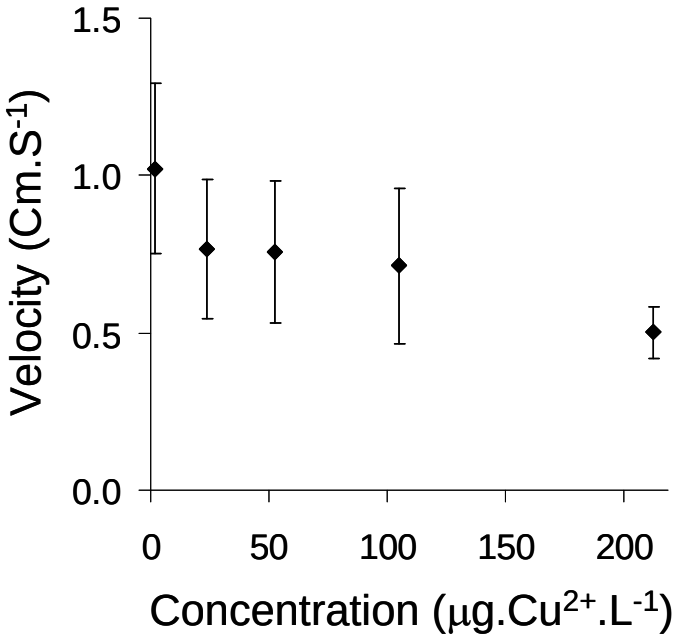
1.5
1.0
0.5
0.0
Velocity (Cm.S-1)
0
50
100
150
200
Concentration (µg.Cu2+.L-1)

Figure 1.

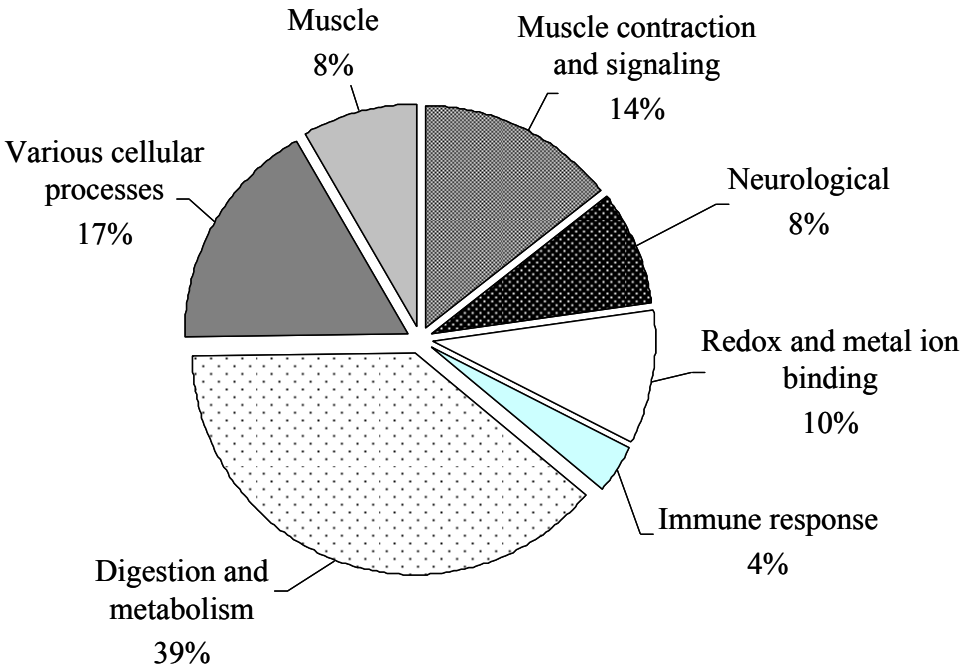
Muscle
8%
Muscle contraction and signaling
14%
Neurological
8%
Redox and metal ion binding
10%
Immune response
4%
Digestion and metabolism
39%
Various cellular processes
17%

Figure 2.

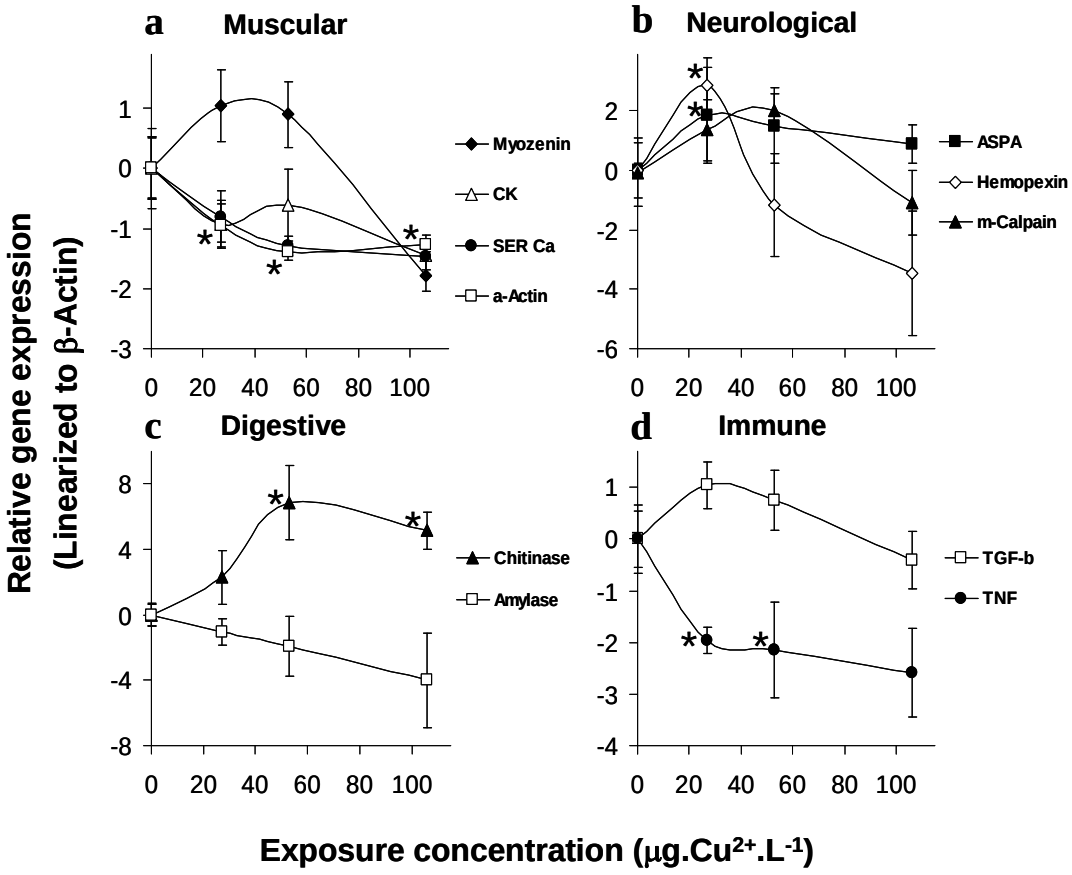
a Muscular
b Neurological
c Digestive
d Immune
Relative gene expression (Linearized to β-Actin)
Exposure concentration (μg.Cu2+.L-1)
Myozenin
CK
SER Ca
a-Actin
ASPA
Hemopexin
m-Calpain
Chitinase
Amylase
TGF-b
TNF

Figure 3.

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Sublethal responses to ammonia in the endangered delta smelt; *Hypomesus transpacificus* (Fam. Osmeridae)

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Abstract:

The delta smelt (*Hypomesus transpacificus*) is a pelagic fish species endemic to the Sacramento-San Joaquin Estuary in Northern California, listed as endangered under both the USA Federal and Californian State Endangered Species Acts and acts as an indicator of ecosystem health in its habitat range. Interrogative tools are required to successfully monitor effects of contaminants upon the delta smelt, and to research potential causes of population decline in this species. We used microarray technology to investigate genome-wide effects in fish exposed to ammonia; one of multiple contaminants arising from wastewater treatment plants and agricultural runoff. A 4-day exposure of 57-day old larvae resulted in a measured un-ionized ammonia (NH_3) LC_{50} of 147$\mu g.L^{-1}$, a NOEC of 66 $\mu g.L^{-1}$ and LOEC 105 $\mu g.L^{-1}$. We assessed genome-wide expression at 105 $\mu g.L^{-1}$ and selected genes were further investigated as molecular biomarkers using quantitative PCR analyses on exposures to 23, 66, 105, 228 and 439 $\mu g.NH_3.L^{-1}$. Genes predominantly encoding for membrane bound proteins responded significantly to ammonia exposure, however, neurological and muscular activity were also impaired. We present here our functional gene classification and further investigations into neurological, muscular, immune, growth and development responses significantly affected by exposure to this contaminant.

Keywords: 'Hypomesus transpacificus', 'delta smelt', microarray, biomarker, ammonia

Introduction.

Contaminants and their potential deleterious effects to fish in the Sacramento-San Joaquin Estuary in Northern California are of particular interest due to negative long-term population trends and a possible step decline in numbers of several pelagic fish species in the years 2000-2001 (Bryant and Souza, 2004; Feyrer et al., 2007; Hieb et al., 2005; Sommer et al., 2007). This trend, known as the pelagic organism decline, has been the focus of an increasing number of investigations over the past several years (Brown et al., 2009; Connon et al., 2009; Sommer et al., 2007). Delta smelt (*Hypomesus transpacificus*) is one of the species of concern. It is endemic to the Delta and has been listed as endangered under both the USA Federal and Californian State Endangered Species Acts.

Ammonia (NH_3) originating from municipal wastewater treatment plants, agricultural activity and numerous other sources, is one of multiple contaminants of concern in delta smelt habitat.
The term ammonia/um refers to two chemical species which are in equilibrium in water (NH_3, un-ionized and NH^{4+}, ionized or nitrogenous ammonia) according to $NH_3 + H^+ \leftrightarrows NH^{4+}$. Tests for ammonia/um usually measure total ammonia plus ammonium, while the toxicity is primarily attributable to the un-ionized form. In general, more un-ionized ammonia and greater toxicity exist at higher pH, because its relative proportion increases with increasing pH according to the following equations (USEPA, 1985):

$$1 / (1 + 10^{pKa-pH}) = \% NH_3$$
$$\text{where: } pKa = 0.0902 + [2729.9/(°C+273.2)]$$

Temperature will affect this equilibrium, but to a far lesser extent than pH. Acute fish toxicity of ammonia decreases with increasing temperature, but toxicity of total ammonia/um shows no correlation with temperature (USEPA, 1999). This is probably due to an increase in the permeability of biological membranes such as gills by a factor of 2-3 for each 10°C increase in water temperature (Eddy et al., 1995).

The Sacramento River drains into delta smelt spawning and larval nursery areas, thus toxicants present in river water could potentially affect early life stages of delta smelt found downstream. Werner et al. (2010), found that ambient ammonia concentrations were greatest in Cache Slough (≤0.025 mg/L nitrogenous ammonia), and near the Sacramento River confluence with the Deep Water Shipping Channel (≤0.021 mg/L nitrogenous ammonia). Ammonia concentrations in the Sacramento River, downstream from the regional wastewater treatment plant were generally lower (≤0.019 mg/L nitrogenous ammonia), likely due to the lower pH of the river water at this location.

Interrogative tools are required to successfully monitor effects of contaminants upon the delta smelt, and to research potential causes of population decline in this species. Microarray gene profiling is a powerful tool for defining genome-wide effects of environmental change on biological function. We have developed a microarray for delta smelt (Connon et al., 2009) and present here the application of this tool to investigate genome-wide effects in delta smelt exposed to ammonia/um. We further assess specific genomic responses utilizing quantitative PCR, within functional gene pathways, and assess the validity of using molecular biomarkers as monitoring tools of individual and population damage.

Materials and Methods.

Test organisms: Delta smelt were obtained from the Fish Conservation and Culture Laboratory (FCCL), UC Davis and maintained for a minimum of 24 hours in experimental conditions prior to test initiation. All experiments and use of test organisms were approved by the UC Davis Institutional Animal Care and Use Committee (Animal Use Protocol for Animal Care and Use #13361). This institution is accredited by the Association for Assessment and Accreditation of Laboratory Animal Care, International (AAALAC) and has an Animal Welfare Assurance on file with the Office of Laboratory Animal Welfare (OLAW). The Assurance Number is A3433-01.

Exposures: Larval delta smelt (57-d old) were exposed for 4 days to 2.5, 5. 10. 20, 40 and 80 $mg.L^{-1}$ ammonium chloride prepared in culture water obtained from the FCCL, concentrations that correspond to 23, 66, 105, 228 and 439 µg NH_3 L^{-1} (un-ionized ammonia). Controls were maintained in culture facility water with specific conductance (SC) of 930 $\mu S.cm^{-1}$ and pH of 7.9. Larvae were acclimated to control water for 24 h prior to test initiation. Replicate experimental treatments (n=4) were initiated with 10 larvae 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% of the water in each replicate container was renewed at test initiation and on day 2. At test end, surviving fish were euthanized with MS-222 (tricaine methanesulfonate, Sigma, St. Louis, MO, USA), rinsed in de-ionized water and snap-frozen in liquid nitrogen and stored at –80°C for subsequent analyses.

Experimental physicochemistry: Water temperature, pH, and DO were measured daily. Conductivity was measured at test initiation. Ammonia nitrogen (NH_4^+-N) concentrations were measured prior to each water renewal and at test termination.

Genomic assessments - microarrays: Development of the delta smelt microarray was described in Connon et al. (2009) (Connon et al., 2009), briefly we utilized a cDNA microarray with 8,448 expressed sequence tags (ESTs) which were pin-printed in duplicate onto epoxysilane coated glass slides. RNA was extracted from frozen whole, individual organisms, using Trizol Reagent (Invitrogen) as per manufacturer's guidelines. cDNA was synthesized from a total of 1ug total RNA, and amplified using a SuperScript^{tm} Indirect RNA Amplification System (Invitrogen) and labeled with and labeled with Alexa fluor dyes (Invitrogen) as per manufacturer's instructions. Microarray assessments were carried out using quadruplicate treatments. Microarray hybridizations were performed using an automated Tecan HS4800 hybridization station. Slides were scanned using a GenePix 4000B scanner (Axon Instruments). Microarray images and data from exposed delta smelt can be accessed at under the pelagic organism decline (POD) section at: http://www.vetmed.ucdavis.edu/apc/WernerLab/subpage/pelagic_organism_decline.html.

Data was analyzed using LIMMA GUI (Linear model for microarray analysis graphical user interface) (Smyth, 2005), written in the R-programming language available through Bioconductor http://www.Bioconductor.org. Data was normalized within arrays using print-tip Lowess and between arrays applying aquantile normalization methods (Livak and Schmittgen, 2001). A linear model fit was computed using the duplicates on the arrays and the least-squares method, no multiple assessment methods were applied to eliminate false positives as our aim was to increase the number of genes available for biomarker assessment, and qualify these through quantitative PCR.

Sequencing of differentially expressed features was carried out at the CA&ES Genomic Facility, UC Davis. Basic Local Alignment Search Tool; translated nucleotide (BLASTx) searches were performed on specific fragments that responded significantly to the exposure treatments. Only genes that were differentially expressed following exposure were sequenced. Sequences were annotated according to homologies to protein database searches using translated nucleotide sequences and direct nucleotide queries (http://blast.ncbi.nlm.nih.gov/Blast.cgi). Sequences were only annotated if they were found to have a BLASTx match with the expect value smaller than $1x10^{-5}$ and a score above 50.

Differentially expressed genes were classified according to the Kyoto Encyclopedia of Genes and genomes (KEGG - http://www.genome.jp/kegg/kegg2.html) and Gene Ontology (GO - http://www.uniprot.org/uniprot), and information gathered from literature, into functional groups. Classification was carried out based on gene expression changes in respect of control subjects, regardless of whether these were up- or downregulated. Specific genes of interest were selected for further investigation using quantitative PCR (see below).

Genomic assessments – qPCR: Genes for quantitative PCR assessments were selected according to level of expression significance, knowledge base from literature, and functional classification. Primer and probes for q-PCR analyses were designed using Roche Universal Probe Library Assay Design Center (https://www.roche-applied-science.com). Designed primers were obtained

from Eurofins MWG Operon (http://www.eurofinsdna.com), and TaqMan probes were supplied by Roche. Sequences for all genes assessed by q-PCR analyses have been submitted to GenBank (http://www.ncbi.nlm.nih.gov). Respective primers and probe systems for investigated biomarkers are detailed in Table 1. Complementary cDNA was synthesized using 1.0 µg total RNA, with random primers and SuperScript® III reverse transcriptase (Invitrogen), and diluted to a total of 120 µl with nuclease free water to generate sufficient template for q-PCR analysis. TaqMan Universal PCR Mastermix (Applied Biosystems) was used in q-PCR amplifications. SDS 2.2.1 software (Applied Biosystems) was used to quantify transcription. We used the geNorm algorithm (Vandesompele et al., 2002) to estimate the variability of the reference genes, and to determine an optimal normalization gene. Quantitative PCR data was analyzed using the relative quantification 2(-Delta Delta CT) method (Livak and Schmittgen, 2001). Expression was calculated relative to β-actin determined by GeNorm as the least variable gene in this study. Quantitative PCR data were not normally distributed, therefore, significant differences in gene expression, relative to the unexposed controls, were assessed using two-tailed Mann-Whitney U test, single comparison alpha = 0.05, with Bonferroni's correction experiment-wide alpha = 0.15, treating each gene as a separate experiment.

Table 1. Primer probe systems designed from microarray assessments on larval delta smelt.

Gene Name	Gene Code	Primer Left	Primer Right	Probe No.
adenylate_kinase	ADK	ctgtcttctggggacctgttg	ctcctttctgcataattgcctgt	36
calmodulin	CaM	ttccttattcgacatggatggc	gcagacccagtgactgcatg	17
claudin-10	CLDN10	ctgcctcggattctttggtg	cctccaattttggtgcacttc	140
epimorphin	EpiM	ctttcgggaaaggaccaaaac	tgcttgtcacttttcccagttatc	94
hla	HLA	atcgtgtctgtggagaaacaggt	ggaagctctggttgaactcgg	25
keratin-15	Ker15	ccagcaaaaccagttactcctcc	cctgatgagcctccatacctca	38
myosin-regulatory-light-chain-2	MYL2	catgggagaccgcttcacc	tgtcgatgggagcttcacg	10
septin-3	SEPT3	ggctttgacctcaacattatggt	cttgagcagagtgttgaccagagt	60
sirtuin-6	SIRT6	gaagccgacaggacgctact	ttccctctgcaggctctgag	1
transmembrane-4-l6-family-member-4	Tm4sf4	ccctggctctcatctccatc	ccatctttggcatacttcacc	64
tropomyosin	TPM	tcccttaacagacgcatccag	cagtagccagacgctcctgtg	101
tubulin-folding-cofactor-b	TBCB	gactcctgcagctggtatgga	ccagcttctgcaggaacttgtc	78
Alpha-Actin	A-Actin	cctgcctcgtcgtactcctg	catcctggcttccctgtcc	11
Amylase	Amy	gatcaccatgttcttgatctgacg	ccatcaatcctgaccaaaacctg	99
Beta-Actin	B-Actin	tgccacaggactccatacc	catcggcaacgagaggtt	12
Creatine Kinase	CK	cgatcggcgttggagatg	gccaagttcaacgagattctgg	163
Myozenin	MyoZ	ccaatgtcgtgctggtacacc	ctgccagacattgatgtagcca	106
SER-Ca	SER CA	catgatcattgggggagca	tgctgtgatgacaacgaggac	148
TGF-B	TGF-b	caacggcatagtgcatgtgg	gaatgtgtgcacgttgttggt	76
Tumor Necrosis Factor	TNF	ctttttccgctgttccatgttc	gttaccagcatacgcagtgtcc	2
Aspartoacylase	ASPA	ggaggcacacatgggaatg	cttcctctgaatctctgttccattatc	109
Hemopexin	HPEX	catgcactacgaggacgacaag	tggtagtagctgaacaccttgctg	143
Titin-a	Titin	tgatcactggcgtgaaagagg	caagctcattggacagtttgagg	159
Zona Pellucida	ZPA	catgcggctgagtttggataa	tgccattgatagcatcaacttca	106

Results and Discussion.

Acute toxicity: 4-day exposure of 57-day old larvae to ammonium chloride resulted in a nominal LC_{50} of 13 $mg.L^{-1}$, a NOEC of 5 $mg.L^{-1}$ and LOEC 10$mg.L^{-1}$, corresponding to measured un-ionized ammonia (NH_3) LC_{50} of 147$\mu g.L^{-1}$, a NOEC of 66 $\mu g.L^{-1}$ and LOEC 105 $\mu g.L^{-1}$ (Table 2 and Fig 1).

Table 2. Ammonium chloride toxicity data on 57-d old larval delta smelt (96-h exposure). Calculated and measured ammonia/um concentrations.

	Survival			
	LC50 (mg/L)		NOEC (mg/L)	LOEC (mg/L)
	Estimate	95% C.I.		
Ammonia Nitrogen (Nominal)	13.0	9.3 - 16.5	5	10
Mean Ammonia Nitrogen (measured)	12.0	8.8 - 15.0	5	9
Mean Unionized Ammonia (measured)	0.147	0.109 - 0.182	0.066	0.105

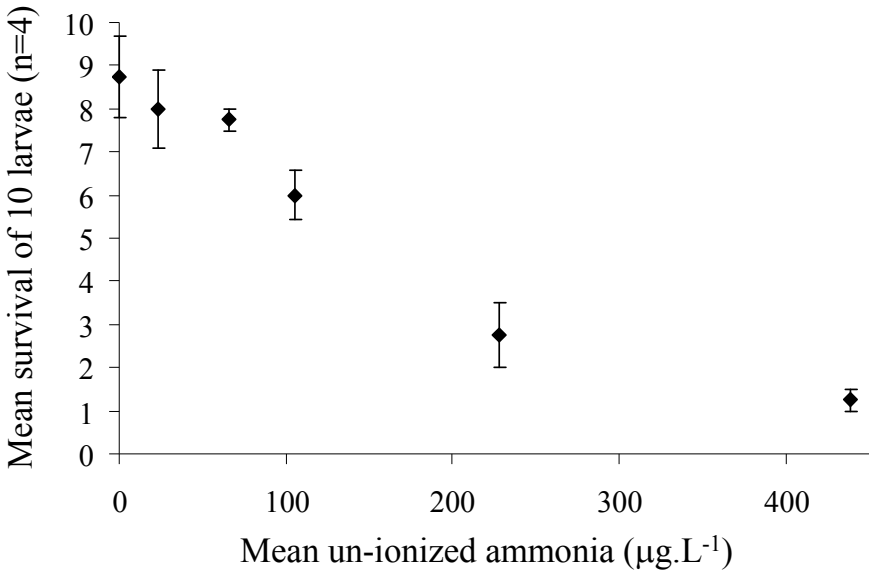

Figure 1. Mean survival (± standard errors) of larval delta smelt exposed to ammonium chloride (96-h exposure). Data expressed as un-ionized ammonia.

Experimental physicochemistry: Temperature, DO and pH remained stable throughout the test duration. Mean data for water temperature, conductivity, DO, pH, ionized and un-ionized ammonia data is shown in table 3.

Table 3. Mean ammonia/um concentrations and physicochemical parameters from 96-h exposure of larval delta smelt to ammonium chloride.

Treatment	Temp (°C)			EC (uS/cm)			DO (mg/L)		
	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*
Hatchery Water	16.5	0.5	8	733	-	1	8.9	0.4	8
2.5 mg/L Ammonia	16.5	0.6	4	748	-	1	9.2	0.7	4
5 mg/L Ammonia	16.5	0.8	4	769	-	1	9.2	0.6	4
10 mg/L Ammonia	16.4	0.7	4	789	-	1	9.1	0.7	4
20 mg/L Ammonia	16.5	0.7	4	847	-	1	9.3	0.3	4
40 mg/L Ammonia	16.5	0.6	4	961	-	1	9.4	0.3	4
80 mg/L Ammonia	15.5	0.3	2	1216	-	1	9.5	0.4	2

Treatment	Ammonia Nitrogen (mg/L)			Un-ionized Ammonia (mg/L)			pH		
	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*
Hatchery Water	0.11	0.05	5	0.001	0.001	5	7.58	0.13	8
2.5 mg/L Ammonia	1.90	0.03	3	0.023	0.009	3	7.64	0.13	4
5 mg/L Ammonia	5.00	0.00	2	0.066	0.034	2	7.62	0.13	4
10 mg/L Ammonia	9.00	0.00	3	0.105	0.033	3	7.62	0.10	4
20 mg/L Ammonia	17.67	0.58	3	0.228	0.120	3	7.63	0.16	4
40 mg/L Ammonia	36.33	2.08	3	0.439	0.247	3	7.59	0.18	4
80 mg/L Ammonia	72.00	5.66	2	0.526	0.133	2	7.47	0.16	2

Genomic assessments: A wide variety of genes from a number of functional pathways were affected by exposure to ammonium chloride (Fig. 2a), of which genes encoding for membrane bound proteins were prominent (56%) (Fig 2b).

Microarray assessment of identified a number of genes that were predominantly related to membrane integrity, membrane bound proteins responsible for ion transport and ionic exchange. This has previously been reported and is attributed to changes in cellular pH resulting from ammonium gradients (Martinelle and Haggstrom, 1993; Randall and Tsui, 2002; Wicks et al., 2002). Neurological and muscular activity was also affected by exposure to ammonium chloride, suggesting possible effects on swimming performance however this was not assessed in this study. Effects of ammonia on swimming performance has been reported in past studies (McKenzie et al., 2009; Wicks et al., 2002).

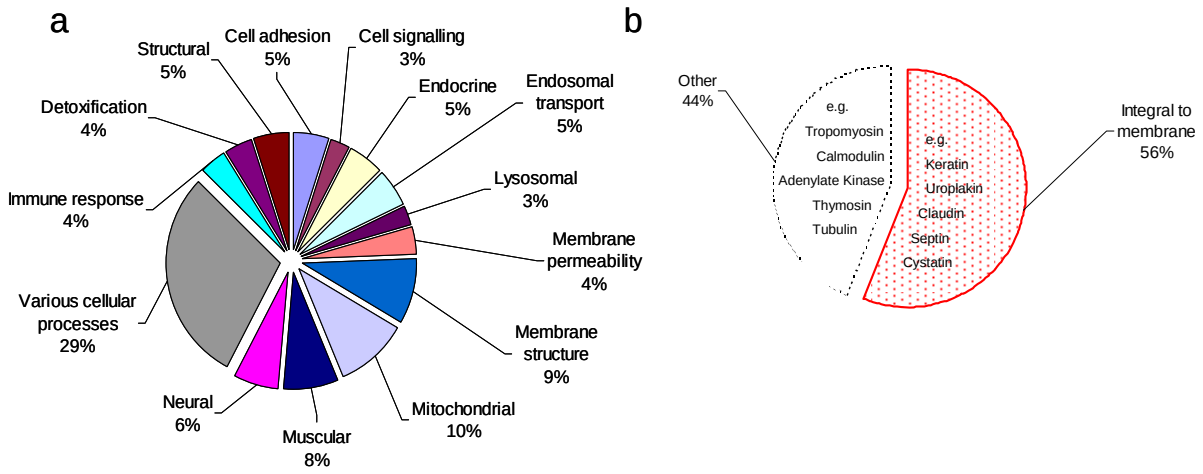

Figure 2. Functional classification of microarray assessed genes responding to 105 µg.L^{-1} ammonium chloride (a) and percentage of genes encoding for membrane proteins (b).

Interestingly, the number, and significance level of genes responding at the assessed concentration (105 µg NH_3 L^{-1}) was low, however the levels of gene expression at this concentration were supported by quantitative PCR assessments. Dose responses of gene expression assessed using qPCR and clustered based on Pearson's correlations of profile similarity, are shown in Figure 3. Responses were predominantly biphasic, suggesting thresholds that correspond with sublethal and acute toxicity.

Biphasic genomic responses measured following exposure to contaminants have been described in detail in a number of studies (Heckmann et al., 2008; Korsloot et al., 2004) and have been postulated to be indicative thresholds of compensatory responses, or tolerance to exposure. The biphasic responses, measured by quantitative PCR, correspond with NOEC and LOEC determined in this study. From a sublethal perspective, that is concentrations at and below NOEC, there is a predominant upregulation of genes concerning membrane proteins (cluster 1), neuromuscular activity (cluster 1 and 2), immune response and digestion (cluster 2), calcium regulation (cluster 4) and of particular interest is Tubulin Cofactor Beta (cluster 5), which has been reported to control directional growth and development of nerve axons (Grynberg et al., 2003; Lopez-Fanarraga et al., 2007). This gene was significantly upregulated in a dose response manner, beyond the biphasic response observed in other gene candidates. Neuromuscular related genes in cluster 3 are highly variable in response, but display an overall downregulation trend.

Effects upon neurological and muscular activity were supported by quantitative PCR assessments. Other studies on larval delta smelt exposed to ammonium chloride, as yet unpublished, have resulted in a decrease in swimming activity. Though not conclusive, the differential responses in creatine kinase, SERCa ATPase and Aspartoacylase could be indicative of swimming performance thresholds. To corroborate this, studies combining genomic assessments and swimming performance would need to be conducted on the same set of organisms. Titin and Tropomyosin were also affected by ammonium chloride exposure, indicating likely effects on muscle structure and development.

Genomic responses at sublethal concentrations of ammonium chloride indicate that membrane systems are being affected by exposure, affecting overall osmoregulation capacity. The biphasic response, observed primarily at 105 μg NH_3 L^{-1} could indicate a threshold beyond which organisms can no longer compensate for exposure.

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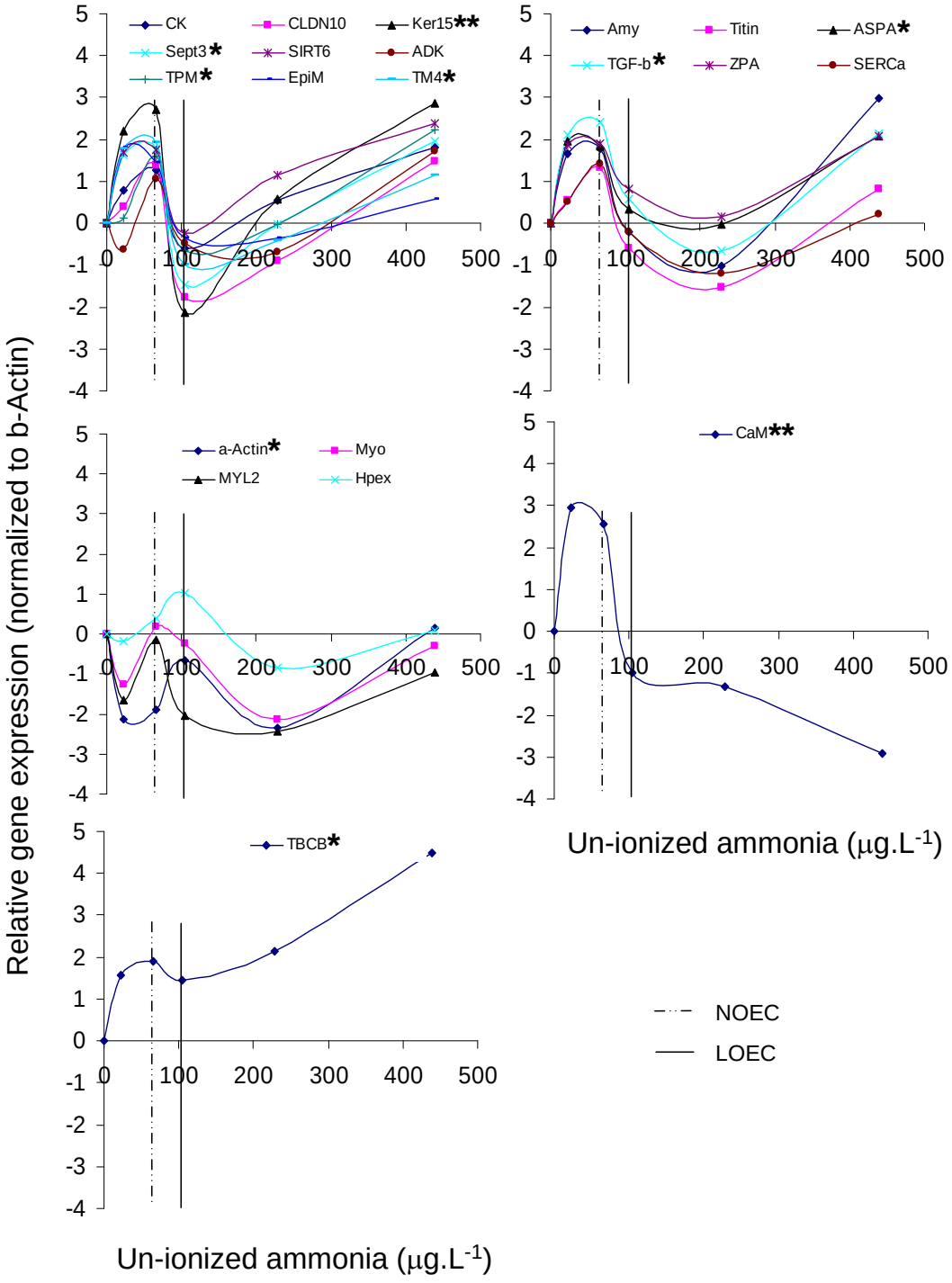
Figure 3. Pearson Correlation cluster analysis of quantitative PCR assessed genes responding in larval delta smelt to 96-h ammonium chloride exposure. Significance levels are displayed for NOEC at 66 µg NH_3 L^{-1} (* = p<0.05 and ** = p<0.01).



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Manuscript *in preparation.*

Cytochrome P450 induction in the striped bass (*Morone saxatilis*): signature biomarkers of past and present xenobiotic exposure

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Introduction

Cytochrome P4501A (CYP1A) is a monooxygenase involved in xenobiotic biotransformation and detoxification. Induction of CYP1A is a powerful biomarker of exposure to polycyclic aromatic hydrocarbons (PAH) and has been widely used in aquatic biomonitoring. Polycyclic aromatic hydrocarbons are widespread contaminants in the San Francisco Bay, a detailed account of which is given by Oros et al (2007). PAHs generally occur as complex mixtures originating from a variety of sources; storm water runoff, wastewater treatment plant effluent, atmospheric deposition and dredged material disposal. Exposure to PAH has been reported to result in weight loss, immune system dysfunction, hyperkeratinization, tumor promotion, edema formation and increased mortality (Poland and Knutson 1982) and alteration of sex hormones in female fish (Spies and Rice Jr 1988).

Along with other pelagic fish species in the San Francisco Estuary, the striped bass (*Morone saxatilis*) have suffered a significant decline in numbers (Bennett and Moyle 1996; Feyrer et al. 2007), and contaminants are likely to have contributed to this decline (Bailey et al. 1994; Bennett et al. 1995; Ostrach et al. 2008; Stevens et al. 1985). Striped bass were introduced from the East coast of USA in the late 1880s, now supports a valuable recreational fishery, and is among the most important piscivorous fish of the Estuary.

The aim of this study was to investigate the dynamics of CYP1A induction at different biological levels of organization (RNA transcription / protein synthesis / catalytic activity) in order to define the respective ranges of utilization as biomarkers of PAHs contamination in this species, and to better understand the relationship of CYP1A gene expression with higher level cellular effects. Toxic responses are directly linked to a contaminant's mode of action, and are generally preceded by alterations in gene transcription (Connon et al. 2009). Although RNA is not the final product of a gene, as the gene function is carried out by the resulting protein, protein expression is influenced and regulated by many processes downstream of mRNA synthesis. Thus by measuring CYP1A mRNA levels it is possible to predict resulting protein activity.

β-naphthoflavone (BNF), a commonly used PAH model compound, was used to induce CYP1A responses in juvenile striped bass. This chemical has been widely used as inducer of genes involved in xenobiotic metabolism; phase I and phase II enzymes. As well as inducing cellular detoxification pathways, BNF has been reported to significantly modulate transcription of genes involved in neuroendocrine control of stress and reproduction (Aluru and Vijayan 2008), thus CYP1A has been proposed as a biomarker of exposure to a number of contaminants (Anderson 2007).

Methods

Fish handling: Striped bass juveniles used in this study were one year old fish produced from a domestic brood stock at the Center for Aquatic Biology and Aquaculture. Fish were moved to the experimental facility two days prior the start of the experiment. They were maintained in circular flow-through tanks (10 fish per tank) at 19 – 20 °C water temperature and under natural photoperiod. Fish were fed daily at 2.5% body weight with #3 Crumbled Salmon / Trout (Silver Cup, Murray, UT 84157).

BNF exposures: Fish were dosed through intra-peritoneal injections in two experiments: (i) a 24h dose response to concentrations ranging from BNF (Sigma-Aldrich Corporation) 0.1 to 50 $mg.kg^{-1}$, and (ii) a time response study at 0 h, 6 h, 12 h, 24 h, 2 d, 4 d and 8 d after exposure to BNF $10mg.kg^{-1}$ and to solvent control. Ten fish were used per time / exposure was used. Fish at 0h received no injection. Corn oil (Sunny Select, Super store industries, Lathrop, CA 95330) was used for BNF dilution and as solvent control. Fish were not fed the day prior the injection. Experiments were carried out from 16/07/08 till 24/07/08. Prior to injection, fish were anesthetized with MS-222 $100mg.L^{-1}$. Fish were sacrificed using an overdose of MS-222. Fish livers were dissected and immediately snap frozen in liquid nitrogen.

Sample preparation: Liver tissue was ground in liquid nitrogen to a fine powder using mortar and pestle and aliquoted into two separate microcentrifuge tubes and then stored at -80°C for subsequent molecular and protein analyzes.

RNA extraction and cDNA synthesis: Total RNA was extracted from powdered liver tissue using RNeasy Mini Kit (Qiagen Inc., Valencia, CA), with on-column DNAse digestion, as per manufactures protocol. Samples were quantified using a NanoDrop spectrophotometer and their quality verified by electrophoresis on 1% agarose gel. 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), 10U RNaseOut (RNase inhibitor), and 1mM dNTPs (all Invitrogen, Carlsbad, CA, USA).

Quantitative PCR: We measured CYP1A gene expression in striped bass using specific probes and primers designed for quantitative PCR (qPCR), as detailed in Table 1. β-actin was used as a housekeeping gene, to which CYP1A expression was normalized. TaqMan Universal PCR Mastermix (Applied Biosystems) was used in cDNA amplifications using an automated fluorometer (ABI PRISM 7900 Sequence Detection System, Applied Biosystems).

Table 1. Primer and Probes for CYP1A and β-actin used for quantitative PCR assessments of gene expression in striped bass.

Gene	Primer	Roche Probe
CYP1A	Forward: GCGGCACAACCCCAGAGTA Reverse: CAGCTTTCATGACGGTGTTGAG	No. 65; CTGGAGGA
β-actin	Forward GCAATGAGAGGTTCCGTTGC Reverse GCAGGACTCCATACCGAGGAA	No. 11; CTTCCAGC

CYP1A Protein quantification: A CYP1A competitive-ELISA for striped bass was developed as described by Tom et al (2002). A brief method description is provided below.

CYP1A protein production: A strain of *Escherichia coli* that contained an antigenic fragment of *Lithognathus mormyrus* CYP1A1 gene was obtained from Dr. Moshe Tom, Israel Oceanographic and Limnological Research, Haifa, Israel. This bacterium was grown over night in Lysogeny Broth with 100 µg/mL ampicillin at 37 ºC overnight to produce a starter culture. This was used to inoculate one liter of Terrific Broth + 50 µg/mL ampicillin + 1.0 mM thiamine at 37 ºC. This was grown for four hours and then heme precursor δ-aminolevulinic acid and inducer isopropyl

β-D-1-thiogalactopyranoside (IPTG) was added to a final concentration of 0.5 mM, and the culture was incubated at 28 ºC for 24 hours. After a first centrifugation at 5,000 x g for 15 minutes in 250ml flasks, bacteria were harvested and then condensed into 50 mL centrifuge tube after a second centrifugation. The cells were resuspended into 30 mL of 0.1 M potassium phosphate buffer + 20% glycerol + 0.1 mM DTT and EDTA (pH 7.4). 0.5 mg/ml of egg white lysozyme was added to the buffer and incubated for one hour at 4 ºC. A nonselective protease inhibitor (Sigma P8340) to a dilution of 1/10,000 and 1 µg/ml DNAaseI was added. The resulted spheroplast suspension was then sonicated with a high powered probe sonicator at 25 Watts, 8 times for 30 seconds while being held on ice and then centrifuged at 30,000 g for 2 hours. The CYP1A containing cytosol was saved and the pellet was discarded.

CYP1A protein purification: The CYP1A containing cytosol was purified on a Ni+ NTA Agarose (Qiagen) column equilibrated with 50 mM potassium phosphate buffer containing 0.2 M NaCl, 20 mM glycine, 20% glycerol and supported with 0.1 mM of both EDTA and DDT running buffer. The poly-His tail that was added on the recombinant protein fragment has a high affinity for the Ni+ ion and allows for the crude extract to pass through and enrich the solution with the protein of interest. The protein was eluted from the column with running buffer containing 50 mM histidine. The fractions of protein were analyzed for total protein with the BioRad DC protein assay, and further analyzed with SDS polyacrylamide gel (InVitrogen NuPAGE) to determine the fractions that had high concentrations of CYP1A. These fractions were pooled, and analyzed with the BioRad DC protein assay to determine total protein content and by SDS gel to determine the CYP1A protein concentration. The results of the SDS gel showed a prominent band at 55kDa matching the molecular weight of the CYP1A protein fragment with a concentration of 0.18 µg/µL in the produced stock.

CYP1A competitive-ELISA optimization: The CYP1A protein fraction was coated on high binding 96 well plates (Costar 3925, Black) at a level of 100, 200 ng/mL CYP1A1 protein fragment in Bicarbonate buffer, pH 9.6. This plate was then blocked with PBS buffer pH 7.5 containing 1% Hammerstein Casein, 1.0 mM Ethylene diamine tetra acetic acid (EDTA), and 0.05% Tween 20. Following this, the plate was first assayed with several concentrations (1/5,000 and 1/10,000 dilutions) of Mouse Anti-Trout CYP1A1 antibody (Biosense Laboratories) and then labeled with a marker antibody, Goat Anti Mouse Horse Radish Peroxidase (HRP) conjugate (Jackson Immuno Research), 1/10,000 dilution. This reporter Ab-Enzyme conjugate catalyzed the reaction with the substrates ADHP or Amplex Red (Anaspec Inc.), 35.4 µM, and Urea Hydrogen Peroxide, 1.76 mM in 0.05 M Sodium Phosphate buffer, pH 7.4. Fluorescence was measured at 544 nm excitation with 590 nm emission on a Wallac Victor 2 Fluorescence Plate Reader. The results confirmed that the antigenic CYP1A protein fragment was present.

The assay has been optimized to contain a 200 ng/ml CYP1A coating step, 1/10,000 dilution of Mouse anti-Striped Bass CYP1A (Biosense Laboratories) competitive binding step, and a 1/10,000 dilution of Goat anti Mouse-HRP reporter Ab-enzyme step of the competitive ELISA assay.

EROD assay for quantification of Cytochrome P450 enzyme activity: Livers were prepared for EROD analysis according modifications of the procedures of (Hodson, Efler et al. 1996; Billiard, Bols et al. 2004). Briefly, livers were homogenized in 10 volumes of ice cold 0.02 M HEPES, 0.15 M KCl, pH 7.5 with 0.22 mM AEBSF and 1/200 Sigma Protease Inhibitor Cocktail.

Following centrifugation at 10,000xG for 15 minutes at 4°C, the S9 supernatant fractions were stored at –80°C. The S9 fractions (40 µl) were then diluted 2-fold and added in triplicate to Costar #3915 black, 96-well plates. Then 40 µl of 10 µM Resorufin ethyl ether in 0.1 M HEPES, 0.1% BSA, pH 7.8 was added for the EROD assays. Forty µl of preincubated NADPH generating system was added to result in a final concentration of 0.8 mm NADP+, 4.8 mM glucose-6-phosphate, 3.2 mM MgCl2 and 0.5 Units/ml glucose-6-Phosphate Dehydrogenase. EROD activity was then determined immediately by measuring fluorescence with Excitation 544 and emission 590 on a Perkin Elmer/Wallace Victor2 fluorescent plate reader. Plates were re-read every 2 minutes and the pmoles product formed estimated from resorufin standard curves. Previous studies (Radenac, Coteir et al. 2004), as well as, preliminary studies in our laboratory, showed that the Resorufin ethyl ether substrate has a slight overlap in fluorescence with that of the resorufin product, and also attenuated the excitation/emission of the product. Thus, resorufin standard curves were spiked on 40 µl of 10 µM substrate to correct for the interference by the initial concentration of substrate. Protein was determined using Bio-Rad DC protein assay with BSA as standard. EROD activity over the 10-minute incubation was calculated as pmole resorufin formed per mg protein per minute.

Results

Gene expression: CYP1A mRNA transcription was significantly upregulated in a dose response manner following 24 h post BNF injection, reaching a plateau at 25 mg.Kg^{-1} at 180-fold change induction ($p<0.001$) (Figure 1). Significant differences were measured at concentrations as low as 0.1 mg.Kg^{-1} resulting in a 12-fold change in gene expression ($p<0.05$). The median response was 10 mg.Kg^{-1}, nearing 100-fold change in gene expression ($p<0.001$), a concentration selected for timed exposure.

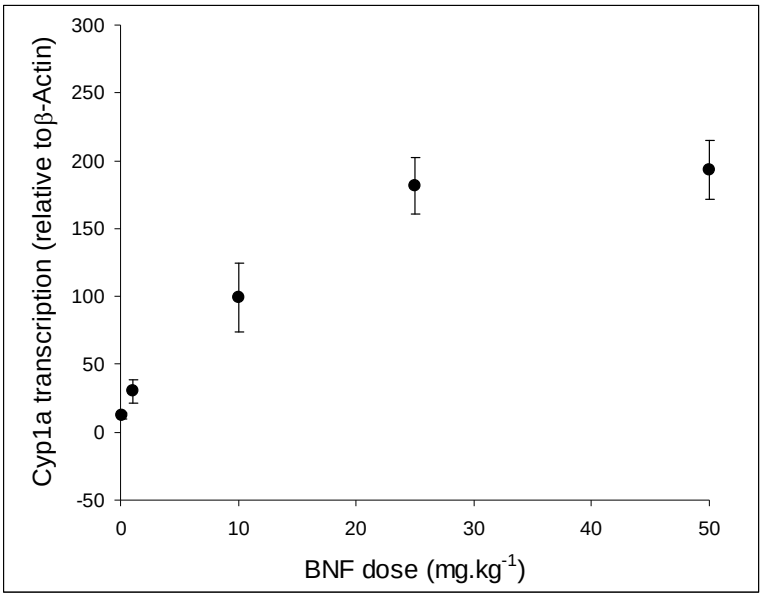

Figure 1. Dose-dependent change in CYP1A gene expression in striped bass exposed for 24 h to 0.1 to 50 mg.Kg^{-1} β-naphthoflavone naphthoflavone (mean +/- standard error).

Timed BNF exposure displayed a 240-fold peak in CYP1A mRNA transcription ($p<0.0001$) at 6 h post injection, reducing after 8 d exposure, though remaining significantly different compared to both time 0, and respective solvent controls ($p<0.001$) at 35-fold change in expression (Figure 2).

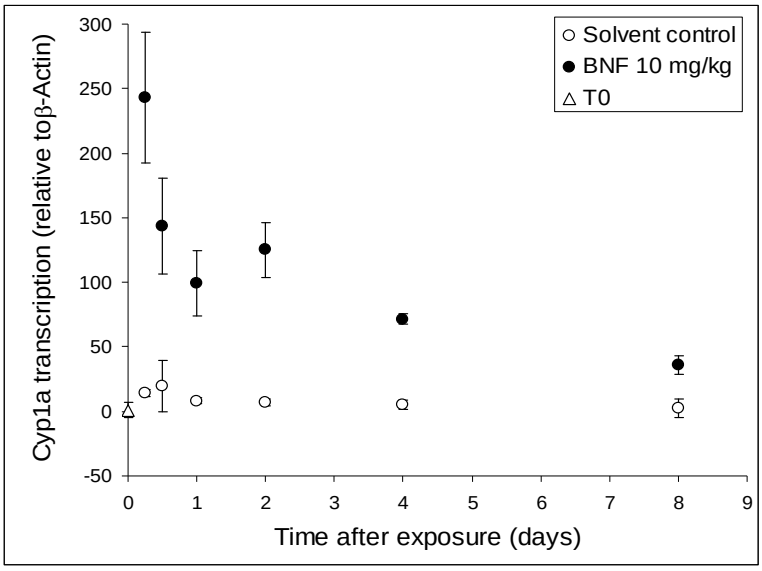

Figure 2. Time-dependent change in CYP1A gene expression in striped bass exposed to 10mg.kg^{-1} β-naphthoflavone (mean +/- standard error).

Protein translation: Quantities of CYP1A protein measured in striped bass exposed to 10 mg.kg-1 β-naphtoflavone increased cumulatively until day 4 of the experiment decreasing slightly by day 8. Protein levels after 8 d were significantly higher (approximately 3–fold) than those measured in control fish (Figure 3).

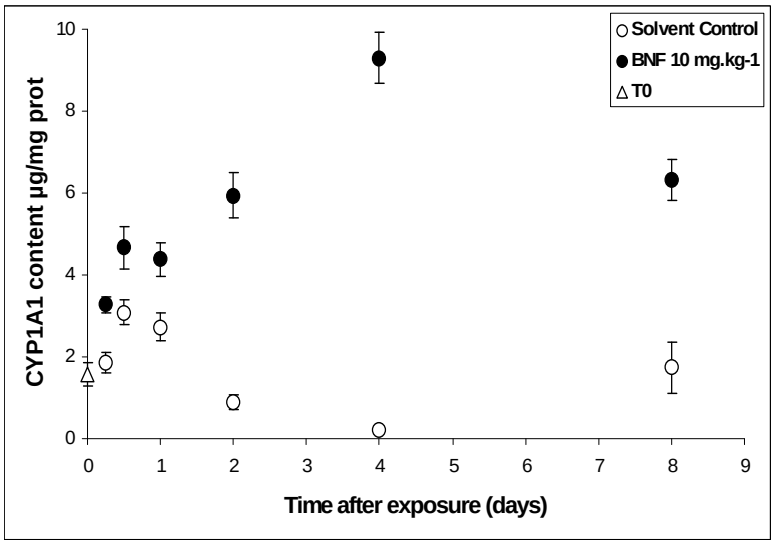

Figure 3. Change in CYP1A protein of striped bass induced with 10 mg.kg-1 β-naphtoflavone compared to solvent control (mean +/- standard error).

Enzyme activity: P450 Enzyme activity, as measured by EROD, increased rapidly upon exposure to BNF, and reached a peak after 12 h post-injection. It declined after that to near control levels by day 8 (Figure 4).

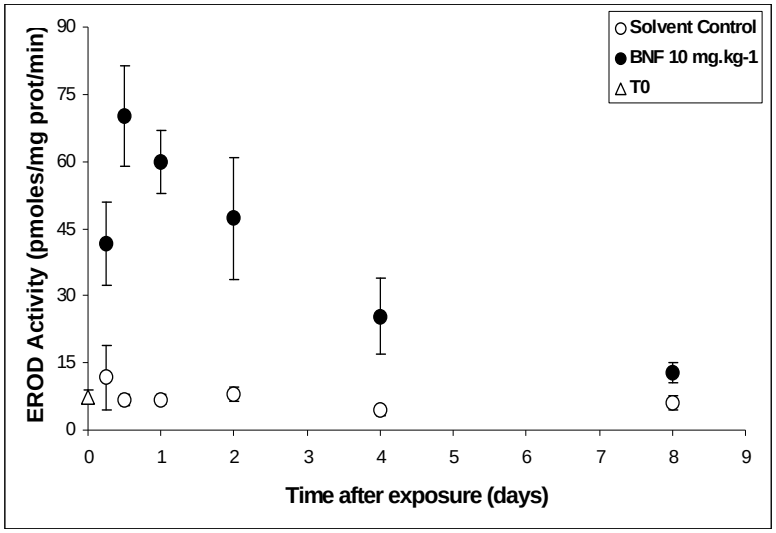

Figure 4. Change in EROD activity of striped bass induced with 10 mg.kg-1 β-naphtoflavone compared to solvent controls (mean +/- standard error).

Comparatively, mRNA transcription and enzymatic activity displayed a similar trend (Figure 5). Expected differences in peak responses were observed, corresponding to mRNA transcription sequentially followed by protein synthesis and resulting enzymatic activity.

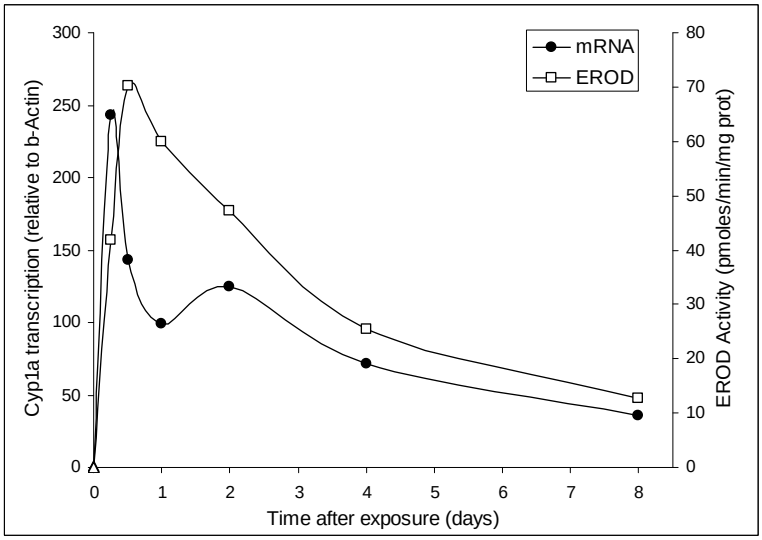

Figure 5. Time dependent change in CYP1A mRNA transcription and EROD activity in striped bass, induced by intraperitoneal injection with 10 mg.kg-1 β-naphtoflavone.

Discussion

All organisms respond to environmental stress by regulating gene expression; templates for protein synthesis. Thus gene expression is the first measurable parameter in response to a stimulus. Linking the molecular, cellular, whole organism and population responses to stressors remains one of the great challenges in ecology and ecotoxicology (Walker et al. 2006), thus the information generated in this study will improve the predictive powers of gene expression measurements as early warning biomarkers of exposure and effect.

CYP1A gene expression responded rapidly to PAH exposure, and corresponding biotransformation and detoxification proteins were synthesized within the first few hours of exposure. An expected lag time was measured between mRNA synthesis (peak at 6h) and resulting protein activity (peak at 12h). Relative lag time is expected to be less than that observed, however interpretation is restricted by the time points measured in the present study.
Accumulation of CYP1A protein and protein activity, measured as EROD, displayed significantly different trends. This may be due to the fact that EROD measures the catalytic activity of CYP1A, specifically involved in O-deetheylisation (Cousinou et al. 2000; Whyte et al. 2000), whilst the competitive ELISA is aimed at quantifying the protein (Brammell et al. 2010; Tom et al. 2002).

CYP1A protein is considered an excellent indicator of PAH exposure, but has also been shown to be involved in neuroendocrine control of stress and reproduction (Aluru and Vijayan 2008), by altering sex hormones (Spies and Rice Jr 1988), as well as affecting the immune system, and promoting tumor formation, and eventual death (Poland and Knutson 1982). The rapid gene expression following BNF injection observed in this study supports the use of this biomarker to detect recent sublethal exposure to PAH. Preliminary results suggest a good correlation between enzymatic activity and gene expression, with a cumulative increase in protein up to day 4 of the experimental period. Measuring both genomic and proteomic responses could thus provide information on the time of xenobiotic exposure, where gene expression is used to detect recent exposures and cumulative protein is indicative of past exposures, whereas the joint response could indicate chronic exposure. Further analyses of data from these tests are currently underway, and additional striped bass genes are being investigated in response to BNF exposure within the context of this study.

Sequential induction of mRNA was followed by protein activity, peaking at 6 and 12 h respectively, and remaining significantly induced over the 8 d treatment. Total CYP1A protein was seen to accumulate throughout the duration of this study, peaking at day 4. This study has demonstrated that all three striped bass biomarker methods used to measure the effect of BNF exposure were successful in detecting CYP1A induction. Though confirmatory studies are still required, models utilizing the differences in responses measured by each of the three biomarkers could be used to detect, in field situations, signatures of past and/or current exposure.

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V.5 Molecular Evaluation of Environmental Contaminant Extracts in Striped bass collected from Semi Permeable Membrane Devices (SPMD) in the San Francisco Estuary.

Connon R.E., D'Abronzo L.S., Ostrach D.J. and Werner I.

Background

Semipermeable membrane devices (SPMDs) are used to assess environmental pollutants from water and air, through the accumulation of hydrophobic organic compounds, such as PCBs, PAHs, and organochlorine pesticides. The principal advantage of SPMD is its sampling of the truly-dissolved and thus bio available phase of these pollutants. SPMDs estimate bioconcentration factors of organic compounds over a period of time, representing a time-weighted average. SPMD derived extracts can be used for conventionally applied aquatic toxicological bioassays.

In an effort to assess bioavailable lipophilic contaminants in the estuary, SPMDs were deployed and extracts used in toxicant bioassays of juvenile striped bass (*Morone saxitilis*). This study was carried out by Dr. David Ostrach. Tissue samples were assessed for gene expression by Dr. Inge Werner's laboratory, in a collaborative approach. Additional tissues will be analyzed in the near future.

Methods

Exposure details:
SPMD extracts dissolved in peanut oil were injected intraperitoneally (100 µL/fish) into 4 and 6 month old hatchery juvenile striped bass in two different exposure experiments. Fish were exposed for 7-days and test terminated by humanely euthanizing the fish in MS222. Livers from each fish were dissected, snap frozen in liquid nitrogen and stored at -80°C for molecular analyses.

RNA extraction and cDNA synthesis
Total RNA from was extracted from liver tissue using a Qiagen RNeasy Mini kit, with on-column DNase digestion following manufacturer's protocols. Complementary DNA (cDNA) was synthesized using 1µg total RNA, with 50 units of Superscript III reverse transcriptase, 600ng random primers, 10 units of RNaseOut, and 1mM dNTPs (all Invitrogen). Reactions were incubated for 50 min at 50ºC, followed by a 5 min denaturation step at 95ºC, and were later diluted 3-fold for subsequent real time - PCR assessments.

Real-time quantitative PCR (rt-qPCR)
Genes investigated in this study were based on sequences, primers and probes previously developed and validated by (Geist et al. 2007), with the addition of □-actin, used as

reference gene, for which primer pairs and fluorescent probes were designed using Roche Applied Science Universal ProbeLibrary Assay Design. All rt-qPCR systems were validated for specificity and amplification efficiencies as described in (Leutenegger et al. 1999). Briefly, a 2-fold dilution series of cDNA samples were tested in triplicate with the respective real-time TaqMan PCR system. The amplification efficiency was calculated using the formula $E=2^{1/S}-1$, where S is the slope curve. All amplification efficiencies were above 90%, validating the specificity of the rt-qPCR systems.

Molecular biomarkers (summarized in table 1) were used to evaluate sublethal stress response of proteotoxicity (HSP70), phase I detoxification mechanism (CYP1a), metal-binding (Metallothionein), endocrine disruption (Vitellogenin) and pathogen-defense (Mx protein).

Gene	Primer Sequences	Roche Probe Number and Sequence
HSP 70	F: CATCCTTTCTGGGGACAAGTCAG R: ACACCTCCAGCGGTCTCAATAC	62 ACCTGCTG
CYP1A1	F: GCGGCACAACCCCAGAGTA R: CAGCTTTCATGACGGTGTTGAG	65 CTGGAGGA
Metallothionein	F: GCGGAGGATCCTGCACTTG R:CAGCCAGAGGCACACTTGGT	68 CTGCTCCT
Vitellogenin	F: CTGATCTGAATTTGGCCTGAGG R: ACCTGTATCCCAAGGACAGTGC	156 GCTGATGG
β-Actin	F: CAATGAGAGGTTCCGTTGC R: CAGGACTCCATACCGAGGAA	11 CTTCCAGC

Table 1. Molecular Biomarkers: List of real-time Quantitative PCR primers and probes used on Striped bass (*Morone saxatilis*)

Real-time TaqMan PCR reactions were prepared with 400nM of each of two primers and 80nM of the appropriate TaqMan probe, and TaqMan Universal PCR Mastermix (Applied Biosystems, Foster City, CA, USA) containing 10mMTris–HCl (pH 8.3), 50mM KCl, 5mM MgCl2, 2.5mM deoxynucleotide triphosphates, 0.625U AmpliTaq Gold DNA polymerase per reaction, 0.25U AmpErase UNG per reaction. A total of 5µl of cDNA was combined with 7µl of the above mix and amplified in 384-well plates with an automated fluorometer (ABI HT 7900 A FAST Sequence Detection System, Applied Biosystems). Amplification conditions were 2 min initial primer annealing at 50ºC and 10 min denaturation at 95ºC, followed by 40 cycles of 15 sec denaturing at 95ºC and 60 sec annealing at 60ºC. SDS 2.2.1 software (Applied Biosystems) was used to quantify product amplification.

Relative quantitation and statistical analyses.
A comparative cycle threshold (*C*T) method as described in (User Bulletin #2, Applied Biosystems) was applied to quantify gene transcription of investigated stress response

genes and values are therefore expressed as relative transcription to □-actin reference gene and *n*-fold transcription relative to oil controls. Both Analysis of Variance (ANOVA) and student-T tests were carried out between SPMD site samples and oil controls, as well as between SPMD dialysis and oil controls. Differences between the two tests dates were also assessed through ANOVA and student t-tests.

Results and Discussion

There were significant temporal variations in gene expression over the four SMPD deployment periods (Figure 1 and 2 – presented separately due to expression scale differences)

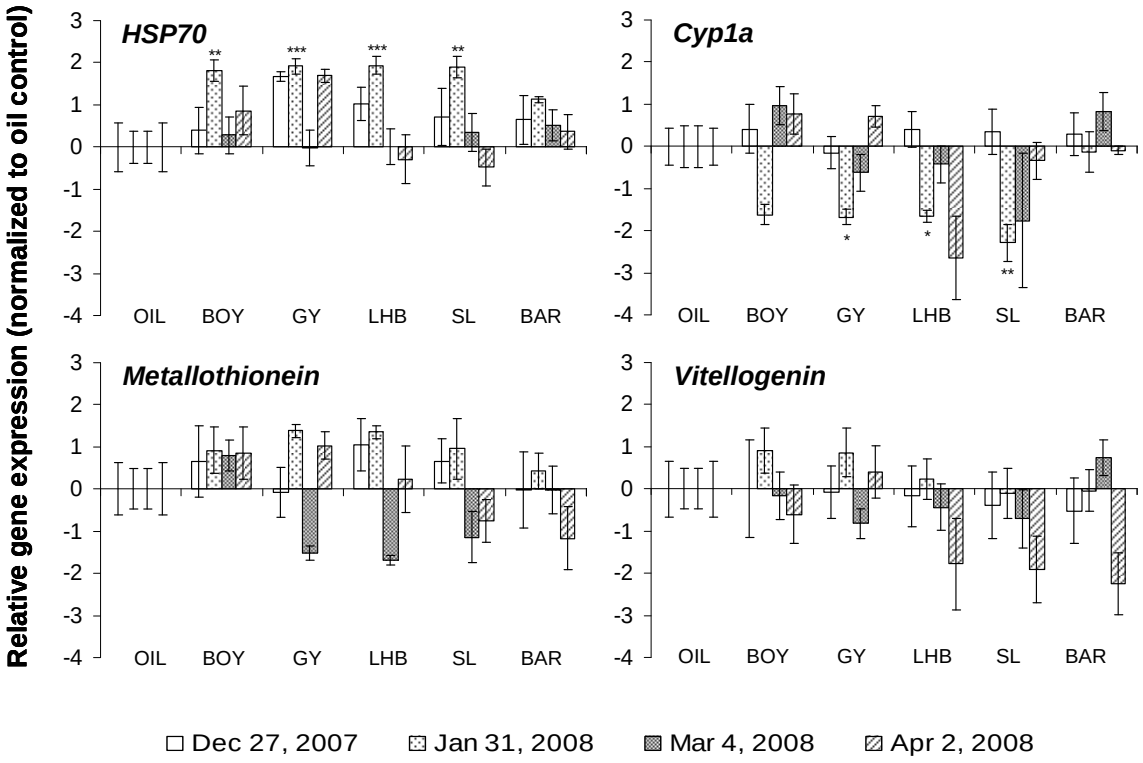

Figure 1. Gene Expression: Biomarker transcription of four selected genes in Striped bass responding to intraperitoneal doses of SPMD accumulated contaminants from five sites in the San Francisco Estuary. Site keys: BAR = Barbie Slough/North Cache Slough; LHB = Little Honker Bay; BY = Boyngton Slough; GY = Goodyear Slough & SL = Sherman Lake. (* $p<0.05$, ** $p<0.01$, ***$p<0.001$).

Heat Shock Proteins (HSP70) were predominantly up-regulated confirming contaminant induced stress, and that protein increase protein synthesis was still induced at the end of the tests. Expression levels were significantly up-regulated at all sites except for Barbie Slough/North Cache Slough (BAR).

CYP1a were predominantly down-regulated at sites Little Honker Bay (LHB) and

Sherman Lake (SL) suggesting probable short term induction leading to sufficient protein synthesis for detoxification purposes. Goodyear Slough (GY), LHB and SL displayed significant down-regulation in respect of oil controls.

Metallothionein displayed both up and down regulations, with temporal variations. Down-regulation, though not significant at test termination, may be indicative of sufficient protein synthesis for metal sequestration at lower doses, whilst mRNA levels were still highly expressed at 48 hour with elevated contaminants.

Interestingly vitellogenin was down-regulated at LHB, SL and BAR at similar time-points in April 2008, though the expression levels were not significantly different to oil controls.

The cytokine encoding for MX protein (presented in fig 2), was significantly up-regulated at Boynton Slough (BOY) and SL, suggesting effects upon the immune system.

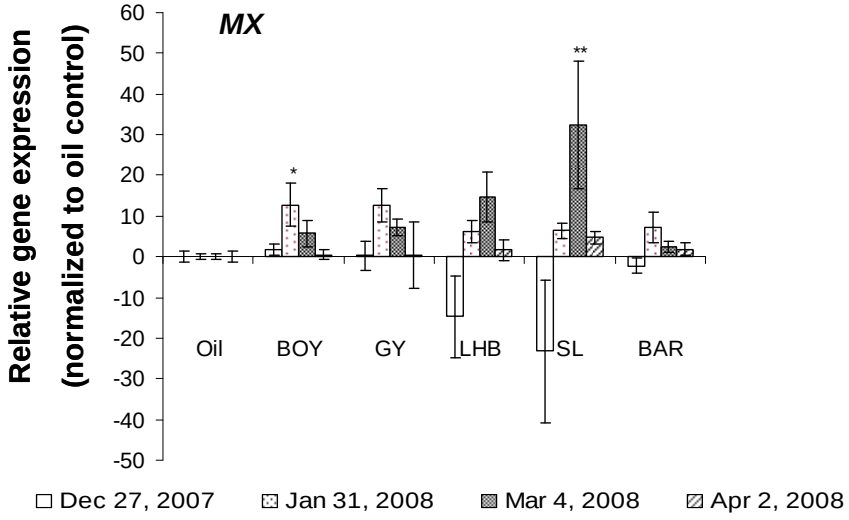

Figure 2. Gene Expression: Biomarker transcription of MX in Striped bass responding to intraperitoneal doses of SPMD accumulated contaminants from five sites in the San Francisco Estuary. Site keys: BAR = Barbie Slough/North Cache Slough; LHB = Little Honker Bay; BY = Boyngton Slough; GY = Goodyear Slough & SL = Sherman Lake. (* $p<0.05$, ** $p<0.01$, ***$p<0.001$).

In summary, HSP70 up-regulation confirms general stress at sites BOYS, GY, LHB and SL, with little to no variation in BAR. Interestingly, the same sites display a down-regulation in Cyp1a, a probable indication that processes have synthesized sufficient protein for this phase I detoxification enzyme. Both BOY and SL samples appear to have further effects upon the striped bass immune system.

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Sublethal toxicity of commercial insecticide formulations and their active ingredients to larval fathead minnow (*Pimephales promelas*)

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ABSTRACT

Toxic effect concentrations of insecticides are generally determined using the technical grade or pure active ingredient. Commercial insecticide formulations, however, contain a significant proportion (>90%) of so-called inert ingredients, which may alter the toxicity of the active ingredient(s). This study compares the sublethal toxicity of two insecticides, the pyrethroid bifenthrin, and the phenylpyrazole fipronil, to their commercial formulations, Talstar® and Termidor®. Both insecticides are used for landscape treatment and structural pest control, and can be transported into surface water bodies via stormwater and irrigation runoff. We used larval fathead minnow (*Pimephales promelas*), to determine effects on growth and swimming performance after short-term (24 h) exposure to sublethal concentrations of pure insecticides and the respective formulations. Significantly enhanced 7 d growth was observed at 10% of the 24 h LC_{10} (53 μg L^{-1}) fipronil. Swimming performance was significantly impaired at 20% of the 24 h LC_{10} (0.14 μg L^{-1}) of bifenthrin and 10% of the 24 h LC_{10} of Talstar® (0.03 μg L^{-1}). Fipronil and Termidor® led to a significant impairment of swimming performance at 142 μg L^{-1} and 148 μg L^{-1} respectively, with more pronounced effects for the formulation. Our data shows that based on dissolved concentrations both formulations were more toxic than the pure active ingredients, suggesting that increased toxicity due to inert ingredients should be considered in risk assessments and regulation of insecticides.

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1. Introduction

Insecticides are contaminating surface water bodies of agricultural areas in California, USA, and elsewhere (Schulz, 2004; TDC-Environmental, 2008; Werner et al., 2004). It is, however, a misconception that attributes insecticide use to agricultural activities alone.

Insecticides are also heavily used in urban areas where application by homeowners and professionals for mosquito control, landscape treatment and structural pest control results in an extensive source of contamination (Budd et al., 2007; Sandahl et al., 2007). Even if not applied in the vicinity of surface water bodies, insecticides can be transported via irrigation runoff and stormwater into urban streams and waterways (Brady et al., 2006; Weston et al., 2005). Aquatic invertebrates and fish thus become targets of toxic substances at potentially hazardous concentrations. This is of special concern if sensitive larval and developmental stages are affected.

Toxicity of insecticides to fish and other aquatic species is generally determined via threshold concentrations such as LC/EC_{50} for the pure active ingredient (A.I.) of commercial products (Cox and Surgan, 2006; USEPA, 2007a). However, commercial products contain the A.I. mixed with non-insecticidal ingredients, so-called “inert” or “other” ingredients, which in some cases comprise more than 90% in volume of insecticide formulations (Cox and Surgan, 2006). They need not be identified on the product label, unless classified as highly toxic (USEPA, 2007b), and act as adjuvants, solvents, emulsifiers, surfactants and/or preservatives. Numerous commercial formulations often exist for each A.I., and it is known that availability and toxicity of the A.I. may be substantially altered by inert ingredients (Schmuck et al., 1994). Studies have shown that in many cases the toxicity of commercial formulations is higher than that of the active ingredient, but this is not always the case. Mayer and Ellersieck (1986) compared the toxicity of 161 technical grade pesticides to their formulations and showed that overall toxicity was not affected in 57%, decreased in 11% and increased in 32% of the cases. In a more recent study (Schmuck et al., 1994), 95% of 273 herbicide, fungicide and insecticide formulations were more toxic to fish than the respective pure A.I. The study presented here aims to contribute information about the comparative toxicity of pure bifenthrin and fipronil and two of their formulation products focusing on sublethal endpoints in larval fish. To our knowledge no such information is currently available for these substances.

Both bifenthrin and fipronil are widely used in structural pest control and other urban and agricultural applications (Oros and Werner, 2005; TDC-Environmental, 2008). The pyrethroid, bifenthrin, is one of the most frequently detected contaminants in surface water

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bodies of areas with urban and agricultural land use (Budd et al., 2007). Similarly, the phenylpyrazole, fipronil, was found to be present in runoff from metropolitan areas throughout the United States (Sprague and Nowell, 2008). Both insecticides are commercially available in a large number of formulated products, generally containing <10% A.I. The bifenthrin formulation; Talstar®, contains 7.9% A.I. as microcapsules (as indicated on product label, 2008), where the insecticide is encased in a coat of "inert" ingredients to ensure its slow release and stabilization (Tsuji, 2001). Termidor®, a fipronil formulation, contains 9.1% A.I. in the form of crystalline particles forming a liquid suspension concentrate (as indicated on product label, 2008). Like all pyrethroids, bifenthrin is highly toxic to fish, interfering with Na^+ channel gating in the nerve cell endings, but other ion-channels such as Cl^- and Ca^{2+} channels can be targeted as well (Burr and Ray, 2004). This leads to continuous neurotransmission, causing hyperexcitability, tremors, convulsions and ultimately death (Bradbury and Coats, 1989; Haya, 1989). Reported LC_{50} values of bifenthrin for fish range from 0.15 μg L^{-1} (rainbow trout, 96 h LC_{50}) to 17.5 μg L^{-1} (sheepshead minnow, 96 h LC_{50}) (Kegley et al., 2008; Werner and Moran, 2008). Runoff from residential areas contained bifenthrin at concentrations of 0.12 μg L^{-1} to 6.12 μg L^{-1}, measured at storm water drainage outflows (L. Oki, UC Davis, personal communication). Fipronil is a "new generation" phenylpyrazole insecticide, whose mode of action differs from organophosphates and pyrethroids, to which numerous insects have developed resistance (Bloomquist, 2003; Soderlund, 2008). Phenylpyrazoles interfere with the function of γ-aminobutyric acid (GABA)-gated Cl^- channels (Cole et al., 1993). In insects and mammals, the behavioral effects of GABA antagonists include hyperactivity, hyperexcitability, and convulsions, which are correlated with increased spontaneous nerve activity (Gunasekara et al., 2007). Fish LC_{50} values have been reported for sheepshead minnow (130 μg L^{-1}, 96 h LC_{50}), bluegill sunfish (83 μg L^{-1}, 96 h LC_{50}) and rainbow trout (100 μg L^{-1}, 96 h LC_{50}) (Gunasekara et al., 2007; Kegley et al., 2008). Concentrations measured in irrigation runoff from residential areas ranged from 0.122 to 10.0 μg L^{-1} (L. Oki, UC Davis, personal communication), and ≤9 μg L^{-1} in surface waters downstream of treated rice fields (Schlenk et al., 2001).

Here we tested the hypothesis that the toxicity of the pure active ingredients, bifenthrin and fipronil, differs from the toxicity of their respective formulations, Talstar® and Termidor®. We used swimming performance and growth as toxicological endpoints in larval fathead minnow (*Pimephales promelas* Rafinesque), and a short exposure period (24 h), to mimic runoff-related pulse exposures (Pick et al., 1984; Werner et al., 2004). Sublethal exposure concentrations were based on previously determined acute LC_{10} values.

2. Materials and methods

2.1. Fish source, acclimation and quality assurance

Fathead minnow larvae were obtained from Aquatox Inc. (Hot Springs, AR, USA) at 7 d post-hatch on the day of arrival. Control water consisted of deionized water, modified with salts to meet USEPA specifications (electric conductivity (EC): 265–293 μS cm^{-1}; hardness: 80–100 as mg $CaCO_3$ L^{-1}; alkalinity: 57–64 as mg $CaCO_3$ L^{-1} (USEPA, 2002a)). Fish were acclimated for a minimum period of 4 h in control water at a temperature of 25 °C. During the acclimation period <1% mortality was observed, and the fish fed and swam normally.

During the project period, routine monthly reference toxicant tests were performed using NaCl to ascertain whether organism response fell within the acceptable range according to USEPA requirements (USEPA, 2002a). Each test consists of a dilution series (5 test concentrations) and a control. All test organisms responded normally (within 95% confidence interval of running mean) and sensitivity was considered typical.

2.2. Insecticide exposure

Reference standard grade bifenthrin [[1α3α(2)]-(±)(2-methyl [1,1′-biphenyl]-3-yl)methyl 3-(2-chloro-3,3,3, trifluoro-1-propenyl)-2,2-dimethylcyclopropanecarboxylate], 99% purity (CAS number 82657-04-3), and fipronil (5-amino-1 [2,6-dichloro-4-(triflouromethyl) phenyl]-4 [(triflouromethyl) sulfinyl]-1H-pyrazole-3-carbonitrile), 98.5% purity (CAS number 120068-37-3) were obtained from ChemService inc. (West Chester, PA, USA). Commercial insecticide formulations Talstar® (US EPA Reg. No. 279-3155; 7.9% bifenthrin per volume; FMC Corporation, Philadelphia, PA, USA) and Termidor® (US EPA Reg. No. 7969-210; 9.1% fipronil per volume; BASF Corporation, Research Triangle Park, NC, USA) were purchased commercially. Bifenthrin consists of 97% *cis*-isomer both in the pure compound and the formulated product. Pure fipronil is a 50:50 racemic mixture, just like its formulation product. All insecticide exposure experiments were conducted at the Aquatic Toxicology Laboratory, School of Veterinary Medicine, University of California Davis.

To determine acute toxic effects on survival, 7 d old larval fish were exposed for 24 h to the following nominal concentrations: 0.75, 1.0, 1.5, 2.0, 3.0 and 4.0 μg L^{-1} bifenthrin, 3.0, 4.0, 4.5, 5.0 and 6.0 μg L^{-1} bifenthrin as amount A.I. in Talstar®, 150, 200, 250, 300, 350 and 400 μg L^{-1} fipronil, and 150, 200, 350, 400 and 450 μg L^{-1} of fipronil as amount A.I. in Termidor®. The exposure concentrations used to determine acute toxicity refer to A.I. concentrations (pure chemical or respective formulation) to ensure direct comparability. For the pure substances we used 1 ml L^{-1} methanol (MeOH) as the solvent carrier and one treatment group containing the same MeOH concentration in control water was added as a solvent control. No solvent carrier was required for the formulations as they are designed to mix with water.

Stock solutions were prepared in MeOH for pure insecticides (2000 mg L^{-1}) and used for both, 24 h LC_{50} determination and sublethal exposure experiments.

Exposure concentrations used for the swimming performance and growth test series were calculated as percentages of the nominal LC_{10} values derived from the acute toxicity tests. For each chemical, treatments consisted of a control, solvent control (pure chemicals only), and 10%, 20%, 33% and 50% of the nominal LC_{10}. Each treatment consisted of 13 replicate 600 ml Pyrex beakers containing 250 ml test solution and 10 fish larvae. Subsequently, we used 9 replicates to determine swimming performance at three different time points and 4 replicates to determine growth.

At test initiation, 10 larvae were transferred from the acclimatization tank to each beaker and exposed for 24 h at a water temperature of 25 °C and a 16:8 light–dark ratio. Test vessels were then manually distributed in a random manner, within the exposure water bath. Fish were not fed during the exposure period.

For the sublethal concentrations, sub-samples of each test solution (1 L) were preserved with dichloromethane (Fisher Scientific, USA) at test initiation, shipped overnight to the California Department of Fish and Game Water Pollution Laboratory (Rancho Cordova, CA, USA), extracted within 24 h of arrival, and analyzed using gas chromatography with mass spectrometry and ion-trap detection. Reporting limits for detection of bifenthrin and fipronil were 0.002 μg L^{-1} (recovery 88.3%) and 0.2 μg L^{-1} (recovery 83.1%), respectively. Talstar® samples were filtered through 0.45 μm glass fiber filter to separate microcapsules from the water phase, and "particulate" and dissolved bifenthrin concentrations were determined. Concentrations for Talstar® are presented as the dissolved fraction. Measured and nominal insecticide concentrations are shown in Table 2.

2.3. 7 d growth

Following the 24 h insecticide exposure, fish were transferred to control water and maintained for 6 days at 25 °C and a 16:8 light:dark photoperiod. During transfer, fish were gently rinsed in control water,

using a fine-meshed sieve and moved to vessels containing control water. From days 2 to 7, approximately 80% of the water was exchanged daily and the number of surviving fish was recorded. Physicochemical variables (pH, dissolved oxygen, temperature, EC) were measured per treatment before and after each water exchange and at test termination. Measurements were conducted on pooled replicates of each treatment. After each water renewal the test beakers were manually distributed in a random manner, throughout the exposure waterbath. Fish were fed *ad libitum* twice a day with newly hatched *Artemia nauplii* (ranging from 30 to 50 individuals). At test termination, surviving fish were euthanized with MS-222 (Tricaine Methanesulfonate, Sigma, St. Louis, MO, USA), then transferred to pre-weighed aluminium weigh boats and dried for 24 h at 100 °C. Dry weight per fish (±0.001 mg) was calculated by measuring whole dry weight divided by the number of fish in each replicate.

2.4. Swimming performance ("one minute racetrack")

Swimming performance was measured at three different time points: (1) Immediately after the 24 h insecticide exposure; (2) after a total of 48 h (24 h recovery in control water), and (3) after a total of 7 days (6 d recovery in control water). At each time point, seven fish per replicate from three replicate beakers per treatment were tested using a circular "racetrack" method (Heath et al., 1993a). This racetrack consisted of a 13 cm diameter Petri dish with an upside-down 8 cm diameter Petri dish centrally placed, divided into 8 sectors by radiating lines drawn on the bottom of the testing dish, and filled with control water to a depth of 1 cm. Fish from pre-selected beakers were transferred individually into the testing device and allowed to acclimate for 1 min. A plastic rod was then used to trigger the fish's escape response by gently touching the tail fin every time the fish stopped moving. Due to possible bias in experimental technique, groups of fish were tested in a random manner, without the experimenter's knowledge of exposure concentration following Heath et al. (1993b). The number of lines or sectors crossed by the fish within 1 min was recorded and used as a measure of swimming performance. Water in the testing device was renewed after testing 7 fish from individual replicates.

2.5. Statistical analysis

We used the Comprehensive Environmental Toxicity Information System (CETIS) by Tidepool Scientific Software (McKinleyville, CA, USA) to calculate nominal effect concentrations for 24 h survival (NOEC, LC50, LC_{10}) based on A.I. Statistical analyses of sublethal endpoints utilized the measured dissolved A.I. concentrations. The Shapiro–Wilk normality test was used to evaluate whether quantitative data met the assumptions of the parametric ANOVA. For multiple comparisons the JMP 7.0 Software by SAS Institute Inc. was used. To evaluate differences between treatments in swimming performance and growth data we used one-way ANOVA and Dunett's multiple comparison post hoc test to compare insecticide treatments to controls and solvent controls. Assumptions of normality and homogeneity of variances were met, except for the highest concentrations, but due to the large differences in swimming performance, the ANOVA is considered to be robust (Underwood, 1997), particularly since the distribution of residuals was unimodal.

3. Results

3.1. Water chemistry

Physicochemical parameters measured at the start and end of the 24 h exposure period were within the acceptable range for the test organism (USEPA, 2002a,b) for all experiments and treatments. The measured mean values (±standard deviation) were pH: 7.51 (±0.19), dissolved oxygen 7. 2 (±0.5) mg L^{-1}, temperature: 23.1 (±0.3) °C, and EC: 278 (±6) µS cm^{-1}.

3.2. Sublethal effects

Individual effects were observed for each substance at concentrations below 50% of the LC_{10}. Concentration levels in the following sections refer to the measured dissolved fractions of A.I., or to percentages of the nominal LC_{10} values determined by initial acute toxicity tests (Table 1).

3.2.1. Swimming performance

3.2.1.1. Bifenthrin. Immediately following the 24 h exposure to pure bifenthrin, the swimming performance of fish from the lowest concentration treatment (0.07 µg L^{-1} or 10% LC_{10}) showed no statistical difference to control or solvent control treatments (Fig. 1). Swimming performance of fish exposed to concentrations ≥0.14 µg L^{-1} (20% LC_{10}, $p<0.001$) was significantly decreased compared to solvent controls. In comparison, exposure to the commercial formulation Talstar® led to decreased swimming performance at ≥0.03 µg L^{-1} bifenthrin (10% LC_{10}, $p<0.001$). After transfer to control water for a 24 h recovery period, swimming performance of exposed fish improved in most insecticide treatments. Fish exposed to bifenthrin concentrations of 0.07–0.14 µg L^{-1} as pure chemical (Fig. 1A), and 0.03–0.05 µg L^{-1} as Talstar® (Fig. 1B) recovered completely. After a recovery period of 6 days, no statistically significant differences between treatments were observed. When comparing dissolved bifenthrin concentrations between pure bifenthrin and Talstar®, the formulation was approximately 5 times more toxic than the pure active ingredient.

3.2.1.2. Fipronil. Swimming performance after 24 h was significantly decreased in fish exposed to concentrations ≥142 µg L^{-1} pure fipronil (20% LC_{10}, $p<0.001$) and ≥148 µg L^{-1} Termidor® (33% LC_{10}, $p<0.01$) (Fig. 2). Although the measured concentrations at this time point are in a similar range, the formulation had a stronger negative impact on swimming at higher concentrations. Fish exposed to 192 µg L^{-1} Termidor® (50% LC_{10}) exhibited statistically significant lower swimming activity than fish exposed to 333 µg L^{-1} fipronil treatment (33% LC_{10}). After 24 h recovery in control water no significant effects on swimming performance were observed in fish exposed to pure fipronil, but after the 6 d recovery period, there was a statistically significant negative effect ($p<0.01$, Fig. 2A). In contrast to the pure fipronil treatments, swimming performance of fish exposed to

Table 1
Acute nominal effect concentrations (mortality) for 7 d old fathead minnow after 24 h exposure to bifenthrin, fipronil and their formulations, Talstar® and Termidor®. Effective concentrations, LC_{50} and LC_{10}. Values in parenthesis represent 95% confidence intervals determined via probit analysis.

Substance	NOEC [µg L^{-1}]	LOEC [µg L^{-1}]	24 h LC_{50} [µg L^{-1}]	24 h LC_{10} [µg L^{-1}]
Fipronil, pure	300	350	398.29 (376.27–438.79)	305.57 (275.56–324.12)
Fipronil formulation	200	350	379.47 (355.13–405.48)	233.01 (201.99–307.94)
Bifenthrin, pure	0.5	1	1.90 (1.69–2.12)	0.92 (0.72–1.09)
Bifenthrin formulation	<3	3	4.85 (4.47–5.34)	2.99 (2.36–3.39)

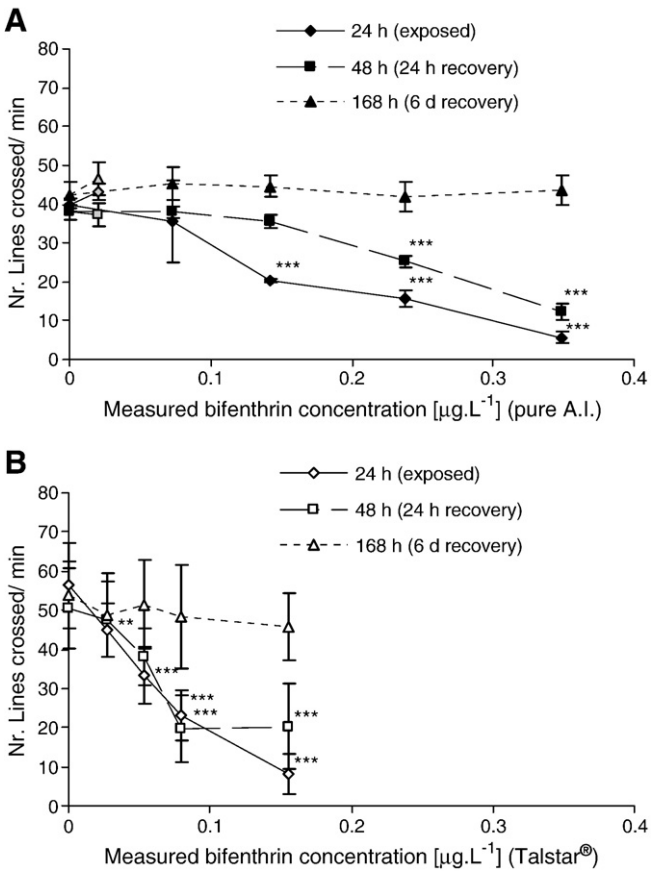

Fig. 1. Swimming performance of larval fathead minnow after 24 h exposure bifenthrin and Talstar®, 24 h recovery and 6 d recovery. Asterisks indicate significant differences in treatments compared to control/solvent control (*: $p<0.05$. **: $p<0.01$. ***: $p<0.001$). Data shown as arithmetic mean ± SD; $n=7$. A: pure bifenthrin, control group shifted to $x=0.02$ for visibility (grey); B: Talstar®.

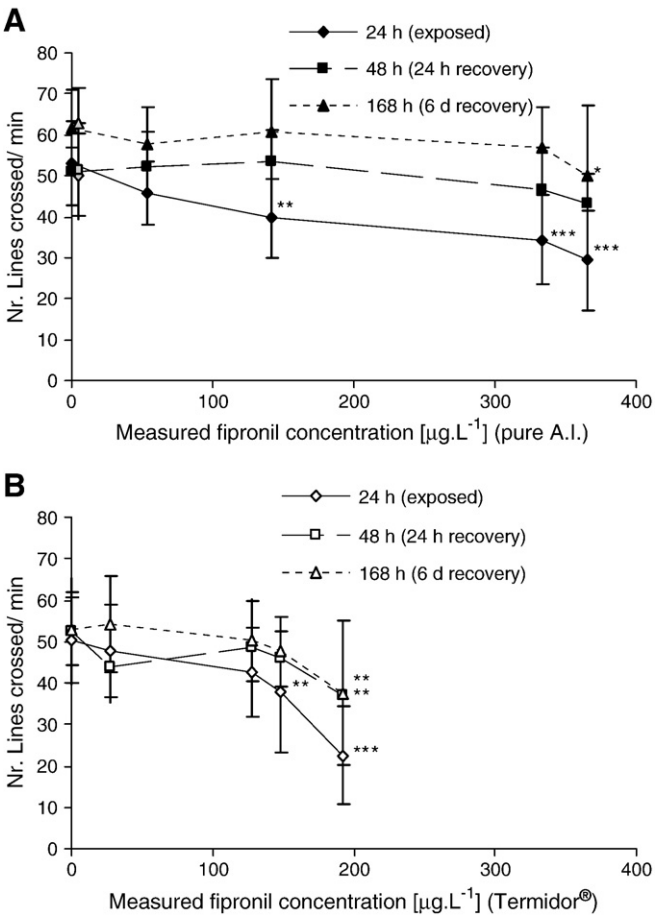

Fig. 2. Swimming performance of larval fathead minnow after 24 h exposure, 24 h recovery and 6 d recovery. Asterisks indicate significant differences in treatments compared to control/solvent control (*: $p<0.05$. **: $p<0.01$. ***: $p<0.001$). Data shown as arithmetic mean ± SD; $n=7$. A: pure fipronil, control group shifted to $x=5$ for visibility (grey); B: Termidor®.

192 µg L^{-1} Termidor® (50% LC_{10}) remained suppressed after the 24 h recovery period. This effect persisted throughout the test, and no recovery of swimming performance was observed after 6 days (Fig. 2B).

3.2.2. 7 d growth

3.2.2.1. Bifenthrin. Neither pure bifenthrin (maximum test concentrations: 0.35 µg L^{-1}, 50% LC_{10},) nor Talstar® (maximum test concentration 0.16 µg L^{-1} A.I., 50% LC10) caused any growth effects in larval fathead minnow (Fig. 3).

3.2.2.2. Fipronil. Fish exposed to pure fipronil at all concentrations tested grew significantly more than fish exposed to the solvent alone (53 µg L^{-1}, $p<0.05$; 333 µg L^{-1}, $p<0.01$; 365 µg L^{-1}, $p<0.01$, Fig. 4). Exposure to Termidor® did not result in negative or positive effects on growth.

In addition to the observed effects on 7 d growth, fish exposed to both pure fipronil and Termidor® showed deformities of the spine (data not presented). Four to five days after the 24 h insecticide exposure, several fish showed scoliosis and in some cases both scoliosis and lordosis. At test termination 5 of the fish exposed to 365 µg L^{-1} and 1 of the fish exposed to 333 µg L^{-1} pure fipronil had developmental abnormalities. The same effect was visible for 4 of the fish exposed to 192 µg L^{-1} and 1 of the fish exposed to 148 µg L^{-1} Termidor®. No such effects were observed in any of the other treatments.

4. Discussion

This study provides new information on the sublethal toxicity of two pure insecticides and two of their commercial formulations to larval stage fathead minnow. Results demonstrate that short-term (24 h) exposures to sublethal concentrations of pure and formulated bifenthrin and fipronil significantly impaired swimming performance

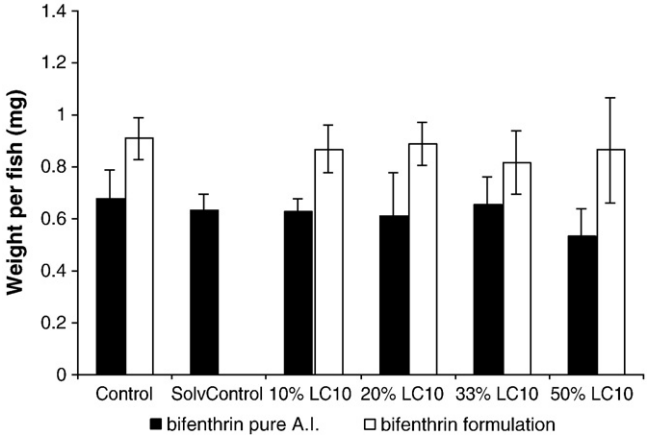

Fig. 3. Average dry weight per fish after 24 h exposure to bifenthrin and Talstar® and 6 d recovery.

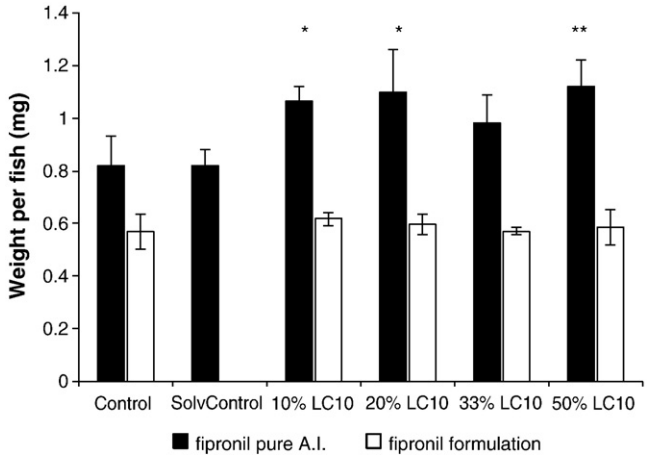

Fig. 4. Average dry weight per fish after 24 h exposure to fipronil and Termidor® and 6 d recovery. Fish exposed to pure fipronil had significantly higher average weight than fish in control treatments (*: $p<0.05$. **: $p<0.01$).

of larval fathead minnows at concentrations as low as 10% of the LC_{10} for the bifenthrin formulation Talstar® and 20% of the LC_{10} for pure bifenthrin.

Bifenthrin and Talstar® concentrations that affected swimming performance (0.14 μg L^{-1} and 0.03 μg L^{-1}, respectively) were in the range of environmental relevance, however, environmental factors such as particulate or dissolved organic matter can reduce bioavailability (Yang et al., 2006) and complicate an ecotoxicological assessment. Sublethal effect concentrations of fipronil and Termidor® (≥142 μg L^{-1} and 148 μg L^{-1}, respectively) were higher than known environmental levels.

Swimming performance is a highly suitable endpoint for estimating individual level effects of environmental contaminants on fish, as it integrates biochemical and physiological processes (Geist et al., 2007; Kane et al., 2005). Especially insecticides with neurotoxic modes of action have been shown to negatively affect swimming ability and predator avoidance (Floyd et al., 2008; Heath et al., 1993b; Little and Finger, 1990). We used a simple and easy to perform test to assess swimming behavior. It simulates predatory chase and integrates both neural and metabolic aspects of fish, since swimming involves nerve cell transmissions and muscle activity (Heath et al., 1993b) which is particularly affected by neurotoxicants (Jin et al., 2009). This is of special ecological importance during early life stages when fish are highly vulnerable to predation. Inability to swim properly after a brief exposure to insecticides therefore negatively affects individual fitness and survival, with potential consequences at the population level (Little and Finger, 1990). As demonstrated in this study fish can recover, but in a field situation, not being able to feed or evade predators for a certain period of time, will likely lead to negative impacts on individual survival and population dynamics.

In this study, growth was not a sensitive endpoint for measuring the effects of bifenthrin. While other pyrethroids have been shown to cause a reduction in growth of fathead minnow and other fish species (Haya, 1989; Jarvinen and Tanner, 1982), we did not observe this effect after bifenthrin and Talstar® exposures. This may be due to the low concentrations used in our experiments (≤50% of the LC_{10}). Floyd et al. (2008) reported significantly reduced 7 d growth in larval fathead minnow after short-term (4 h) exposure to the pyrethroid esfenvalerate, however, effect concentrations were ≥22% of the LC_{50}. The relatively long recovery period (6 d after 24 h exposure) from pyrethroid poisoning may have enabled the fish to compensate for any initial impairment. We did not rigorously quantify food uptake in this study, but during daily water renewal, remaining food quantity was observed to be greater in treatments with decreased swimming performance than in control treatments up to 2 d after insecticide exposure.

Exposure to pure fipronil enhanced growth of larval fathead minnow, while its formulated form, Termidor® did not produce this effect. Enhanced growth following exposure to fipronil has not been previously reported and causative factors should be investigated in more detail, but were beyond the scope of this investigation. A limited number of studies found fipronil to be altering normal thyroid function and thyroid hormone levels in rats (Hurley et al., 1998; Leghait et al., 2009) and chicken (Russ, 2005). As thyroid hormones also play a role in larval and juvenile development of fish (Power et al., 2001) the observed growth abnormalities may be related to this effect.

Developmental effects such as those observed in this study for a small number of the fish exposed to ≥148 μg L^{-1} Termidor® and ≥333 μg L^{-1} pure fipronil, were also reported by Stehr et al. (2006), in particular notochord degeneration and shortening along the rostral-caudal body axis in zebrafish (*Danio rerio*) embryos after continuous exposure to fipronil at nominal concentrations at or above 0.7 mM (333 mg L^{-1}). These authors also observed ineffective tail flips and uncoordinated muscle contractions in response to touch. Although most concentrations used in our study were below that range, similar behavioral abnormalities were observed and resulted in a measurable decrease of swimming performance.

We found strong differences in toxicity between pure and formulated insecticides. Both formulated products were more toxic than the respective A.I., based on measured dissolved concentrations. Talstar® impaired fathead minnow swimming performance at approximately one fifth of the effect concentration of pure bifenthrin. However, when adding the concentration of bifenthrin measured in the particulate fraction of Talstar®, the total concentration that caused negative effects on swimming was approximately 2 times higher for Talstar® than for pure bifenthrin (Table 2). Microcapsules may have been ingested by the larval fish, thus adding a dietary

Table 2
Nominal and measured concentrations for 24 h exposure of 7 d old fathead minnow to bifenthrin, Talstar®, fipronil and Termidor®. Treatment concentrations used for swimming performance and growth tests, calculated as percentages of the LC_{10} value (10%, 20%, 33% and 50% LC_{10}).

Substance	Concentration [μg L^{-1}]	10% LC_{10}	20% LC_{10}	33% LC_{10}	50% LC_{10}
Bifenthrin, pure A.I.	*Measured*	*0.07*	*0.14*	*0.24*	*0.35*
	Nominal	0.09	0.18	0.31	0.46
Bifenthrin, Talstar®	*Measured–dissolved*	*0.03*	*0.05*	*0.08*	*0.16*
	Measured particulate	0.19	0.39	0.57	0.81
	Nominal	0.29	0.59	0.99	1.49
Fipronil, pure A.I.	*Measured*	*53*	*142*	*333*	*365*
	Nominal	31	61	102	153
Fipronil, Termidor®	*Measured*	*28*	*128*	*148*	*192*
	Nominal	23	47	78	117

exposure route to the aqueous exposure to dissolved bifenthrin, which could account for the higher toxicity of the formulated product based on dissolved concentrations. In addition, it is possible that the presence of 0.1 methanol added as a carrier increased bioavailability and toxicity of the pure insecticides, however, we found no difference in swimming performance or growth between control and solvent control treatments. For pure fipronil and Termidor®, effect concentrations for swimming performance were similar, but impairment was more persistent in fish exposed to the formulated product.

Insecticide formulations can act as mixtures and environmental risks cannot be determined by assessing the toxicity of the A.I. alone. The relevance of these findings is obvious as pure insecticides are never applied in the environment. Extrapolating our laboratory results to a field exposure scenario is, however, beyond the scope of this study. For determination of toxicity under environmental conditions many additional factors have to be taken into account. Sediment particles, dissolved organic carbon, water pH and temperature can change the bioavailability and therefore toxicity of pesticides (Maul et al., 2008; Yang et al., 2007). Despite that, the consideration of short-term exposures, delayed effects and sublethal toxicity is of importance as exposure of aquatic organisms to insecticides is most likely to be of short duration and below lethal levels. For example, Brady et al. (2006) demonstrated that the majority of insecticide runoff of two insecticides, diazinon and esfenvalerate, occurred within the first hour of a simulated rain event.

Information on inert ingredients is largely treated as trade secret, but these chemicals have been shown to exert additive or synergistic toxicity, due to either their mechanism of action or through increasing the bioavailability of the A.I. Emulsifiable formulations of pyrethroids were found to be 2.2 to 8.5 times more lethal to fish than the pure substance (Haya, 1989) as a consequence of enhanced uptake via the gill epithelium. In other products, enzyme altering synergists like piperonyl butoxide (PBO) are added (Amweg and Weston, 2007) to enhance toxicity of the A.I. The solvent propylene glycol is part of the Talstar® formulation, but its toxicity to fish is low (fathead minnow 48 h LC_{50}: 790,000 μg L^{-1} (Kegley et al., 2008; TDC-Environmental, 2008)), and it was found to not significantly modify the toxicity of bifenthrin to cultured human cells (Skandrani et al., 2006). Chemicals used in pesticide formulations may also increase mobility of the A.I. thus facilitating pesticide movement into aquatic environments. Suspension liquids such as Termidor® or microencapsulated products like Talstar® are designed to not immediately bind to porous surfaces, and are therefore more susceptible to runoff or leaching. For example, Armbrust and Peeler (2002) reported that the concentration of the insecticide imidacloprid was higher in runoff from turf that was treated with granules than turf treated with wettable powder. Similar formulation effects were observed for herbicide runoff from container plant nurseries (Briggs et al., 2002). Kenimer et al. (1997) reported higher surface runoff of alachlor microencapsulated formulation compared to alachlor emulsifiable concentrate formulation, as microcapsule movement was similar to that of eroded sediment.

Talstar® is formulated as a so-called microencapsulation of bifenthrin, resulting in μm-sized particles, where the A.I. forms a core that is coated by an outer wall consisting of “inert” ingredients (Scher et al., 1998; Tsuji, 2001). The toxicity of this formulation is therefore dependent on how fast and how much of the active ingredient is released through the capsule (Jarvinen and Tanner, 1982). As this formulation is designed to be more persistent at the site of application, the release is probably slow. This explains why measured concentrations of dissolved bifenthrin were lower in the Talstar® experiment than in the exposures to pure bifenthrin. The use of such controlled-release insecticides may lead to lower exposure concentrations but increased exposure time of non-target organisms. Future investigations on these types of products should therefore consider a long-term exposure scenario to lower concentrations.

5. Conclusions

Our study demonstrated that formulated products of two widely used insecticides, the pyrethroid bifenthrin and the phenylpyrazole, fipronil, were approximately 5 and 2 times more toxic to larval fathead minnow than the active ingredients alone. Growth was not a sensitive toxicity endpoint, but the fish's ability to swim normally was impaired at Talstar® (bifenthrin) and Termidor® (fipronil) concentrations 10 and 3 times lower, respectively, than the 24 h LC_{10}. Results suggest that these neurotoxic insecticides can decrease ecological fitness of sensitive aquatic species at concentrations far below the lethal level. We have demonstrated that behavioral endpoints such as swimming are valuable tools to detect sublethal effects of neurotoxic chemicals. Future risk assessments should include information on sublethal endpoints such as swimming behavior, and additional safety factors to account for the greater toxicity of formulated pesticide products.

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Manuscript *in preparation*

Changes in gene expression in larval fathead minnow (*Pimephales promelas*) following short-term exposure to two insecticides, bifenthrin and fipronil.

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Introduction

For environmental risk assessment in the field of ecotoxicological research, molecular methods are becoming more and more important. So called "omics" technologies allow investigation of chemical modes of action from a mechanistic point of view. These techniques are suitable to describe the effects of environmental stressors, including chemical contaminants, on organisms at concentration levels far below the lethal range. This goes hand in hand with ecological relevance, as many chemicals occur at concentrations far below lethal levels. Sublethal concentrations are known to affect organisms at the molecular and individual level that can result in negative consequences at the population and finally ecosystem levels. This is of special concern with regard to sensitive non-target organisms. The application of insecticides in urban and agricultural areas and their effects on aquatic ecosystems are a good example for this. Molecular approaches provide tools for investigating the pathways of adverse effects in the organism. Molecular biomarkers can be rapid, cost-effective and sensitive diagnostic tools for physiological organism impacts.

To study gene responses of fish to insecticide exposure, we used the common model organism, the fathead minnow (*Pimephales promelas*). Effects were determined by quantifying the expression of a suite of biomarker genes involved in molecular pathways involved in organism growth, energy metabolism, muscular and neuronal function and general stress responses. In combination with previously measured effects on swimming performance and growth we evaluated, how useful a small set of biomarkers can be to predict and describe toxic effects of two different insecticides on larval fish.

We exposed fish to two insecticides, the synthetic pyrethroid bifenthrin, and the phenylpyrazole fipronil, both widely applied in urban and agricultural areas. These insecticides are heavily used in urban areas, where application by homeowners and professionals for mosquito control, landscape treatment and structural pest control results in an extensive source of contamination (Budd et al., 2007; Sandahl et al., 2007). Even if not applied in the vicinity of surface water bodies, insecticides can be transported via irrigation runoff and stormwater into urban streams and waterways (Weston et al., 2005). Aquatic invertebrates and sensitive life stages of fish thus become targets of toxic substances at potentially hazardous concentrations.

Both insecticides frequently been detected in surface waters and sediments in California surface waters (Budd et al., 2007, Sprague and Nowell, 2008). Runoff from residential areas contained bifenthrin at concentrations of 0.12 $\mu g.L^{-1}$ to 6.12 $\mu g.L^{-1}$, measured at the the point of storm water drainage outflows (L.Oki, UC Davis, personal communication). Like all pyrethroids, bifenthrin is highly toxic to aquatic organisms. The main mode of action is the interference with Na^+ channel gating in the nerve cell endings, but other ion-channels such as Cl^- and Ca^{2+} channels can be targeted as well (Burr and Ray, 2004). This leads to continuous neurotransmission, causing hyperexcitability, tremors, convulsions and ultimately death (Bradbury and Coats, 1989; Haya, 1989). Reported LC_{50} values of bifenthrin for fish range from 0.15 $\mu g.L^{-1}$ to 17.5 $\mu g.L^{-1}$ (Kegley, 2008; Werner and Moran, 2008). Fipronil is a "new generation" phenylpyrazole insecticide, whose mode of action differs from organophosphates and pyrethroids, to which numerous insects have developed resistance. Phenylpyrazoles interfere with the function of γ-aminobutyric acid (GABA)–gated Cl^- channels (Cole et al., 1993). GABA is a major inhibitory neurotransmitter in the vertebrate central nervous system. In insects and mammals, the behavioral effects of GABA antagonists include hyperactivity, hyperexcitability, and convulsions, which are correlated with increased spontaneous nerve

activity (Gunasekara et al., 2007). Fish LC_{50} values have been reported for sheepshead minnow (130 $\mu g.L^{-1}$), bluegill sunfish (54 $\mu g.L^{-1}$) and rainbow trout (128 $\mu g.L^{-1}$) (Gunasekara et al., 2007, Kegley, 2008). Concentrations measured in irrigation runoff from residential areas ranged from 0.122 to 10.0 $\mu g.L^{-1}$ (L.Oki, UC Davis, personal communication), and ≥9 $\mu g.L^{-1}$ in surface waters downstream of treated rice fields (Schlenk et al., 2001).

Material and Methods

Fish source, acclimation and quality assurance: Fathead minnow larvae were obtained from Aquatox Inc. (Hot Springs, AR, USA) at 7 d post-hatch on the day of arrival. Control water consisted of deionized water, modified with salts to meet US EPA specifications (electric conductivity (EC): 265 - 293 $\mu S.cm^{-1}$; hardness: 80-100 as mg $CaCO_3.L^{-1}$; alkalinity: 57-64 as mg $CaCO_3.L^{-1}$ (US EPA, 2002a). Fish were acclimated for a minimum period of 4 h in control water at a temperature of 25˚C. During the acclimation period <1% mortality was observed, and the fish fed and swam normally.

During the project period, routine monthly reference toxicant tests were performed using NaCl to ascertain whether organism response fell within the acceptable range according to USEPA requirements (USEPA, 2002a). Each test consists of a dilution series (5 test concentrations) and a control. All test organisms responded normally (within 95% confidence interval of running mean) and sensitivity was considered typical.

Insecticide exposure: Reference standard grade bifenthrin [[1α3α(2)]-(±)(2-methyl[1,1'-biphenyl]-3-yl)methyl 3-(2-chloro-3,3,3, trifluoro-1-propenyl)-2,2-dimethylcyclopropane-carboxylate], 99% purity (CAS number 82657-04-3), and fipronil (5-amino-1 [2,6-dichloro-4-(triflouromethyl) phenyl]-4 [(triflouromethyl) sulfinyl]- 1H-pyrazole-3-carbonitrile), 98.5% purity (CAS number 120068-37-3) were obtained from ChemService inc. (West Chester, PA, USA). Bifenthrin consists of 97% cis-isomer. Pure fipronil is a 50:50 racemic mixture.
All insecticide exposure experiments were conducted at the Aquatic Toxicology Laboratory, School of Veterinary Medicine, University of California Davis.

To determine acute toxic effects on survival, 7 d old larval fish were exposed for 24 h to the following nominal concentrations: 0.75, 1.0, 1.5, 2.0, 3.0 and 4.0 $\mu g.L^{-1}$ bifenthrin and 150, 200, 250, 300, 350 and 400 $\mu g.L^{-1}$ fipronil. We used 1 $ml.L^{-1}$ methanol (MeOH) as the solvent carrier and one treatment group containing the same MeOH concentration in control water was added as a solvent control. Stock solutions were prepared in MeOH for pure insecticides (2000 $mg.L^{-1}$) and used for both, 24 h LC_{50} determination and sublethal exposure experiments.

Exposure concentrations used for assessment of sublethal stress responses were calculated as percentages of the nominal LC_{10} values derived from the acute toxicity tests. For each chemical, treatments consisted of a control, solvent control and 10%, 20%, 33% and 50% of the nominal LC_{10} (table 1). Each treatment consisted of 9 replicate 600 ml Pyrex beaker containing 250 ml test solution and 10 fish larvae. We used 3 replicates to sample fish for gene expression analysis at each of three different time points.

At test initiation, 10 larvae were transferred from the acclimatization tank to each beaker and exposed to test solutions for 24 h at a water temperature of 25˚C and a 16:8 light-dark ratio. Test vessels were then manually distributed in a random manner, within the exposure water bath. Fish were not fed during the exposure period. For measuring exposure concentrations, sub-samples of each test solution (1 L) were preserved with dichloromethane (Fisher

Scientific, USA) at test initiation, shipped overnight to the California Department of Fish and Game Water Pollution Laboratory (Rancho Cordova, CA, USA), extracted within 24 h of arrival, and analyzed using gas chromatography with mass spectrometry and ion-trap detection. Reporting limits for detection of bifenthrin and fipronil were 0.002 $\mu g.L^{-1}$ (recovery 88.3 %) and 0.2 $\mu g.L^{-1}$ (recovery 83.1 %), respectively.

After 24 h, a subset of fish from three replicate exposure containers per treatment was sampled. The remaining fish were transferred to control water and maintained according to standard US EPA protocols. Additional samples (3 replicates per treatment) were taken after 48 h, including a 24 h recovery period in control water, and after 7 days, including a 6 d recovery period. Fish were euthanized in MS-222, individually transferred into 1.5 ml Eppendorf tubes and immediately flash-frozen in liquid nitrogen. Samples were stored at -80°C until processing.

Sample processing: RNA was extracted from whole, individual organisms, using QIAGEN RNeasy MiniKit according to manufacturer's instructions. Total RNA concentration was estimated from absorbance at 260 nm (NanoDrop ND 1000) and RNA quality was verified by electrophoresis on ethidium bromide-stained 1.5% agarose gels and A_{260}/A_{280} ratios. 1.0 µg of total RNA was used for cDNA synthesis. Primer and probes for qPCR analyses were designed using Roche Universal Probe Library Assay Design Center (https://www.roche-applied-science.com). Designed primers were obtained from Eurofins MWG Operon (http://www.eurofinsdna.com), and TaqMan probes were supplied by Roche. Primers and probes for investigated biomarkers are detailed in table 1.

Quantitative PCR: Complementary DNA (cDNA) was synthesized using 1.0 µg total RNA, with random primers and SuperScript® III reverse transcriptase (Invitrogen), and diluted to a total of 150 µl with nuclease free water to generate sufficient template for qPCR analysis. TaqMan Universal PCR Mastermix (Applied Biosystems) was used in q-PCR amplifications in a reaction containing 10 mM Tris–HCl (pH 8.3), 50 mM KCl, 5 mM MgCl2, 2.5 mM deoxynucleotide triphosphates, 0.625U AmpliTaq Gold DNA polymerase per reaction, 0.25U AmpErase UNG per reaction and 5 µL of cDNA sample in a final volume of 12 µL.

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 60s 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, CT). SDS 2.2.1 software (Applied Biosystems) was used to quantify transcription

Table 1: Primer pairs and TaqMan probes used in qPCR assessment of molecular biomarkers.

Accession No.	Gene		Primer Sequences	Probe No.
AY538777	Heat shock cognate 70 kDa protein (HSP70)	Left:	ccgttgaagatgagaaactcgc	101
		Right:	cctcattgcacttgtcgagga	
EF628371	Ferritin heavy chain mRNA	Left:	cttgctggttttgccaagttct	63
		Right:	gcccctcttgttctgaaactcc	
EU332792	CYP3A126 (CYP3A126)	Left:	caacccagaggccatgaaga	63
		Right:	gggccttatttgggaaggtct	
-	Parvalbumin	Left:	gctctgtctgctgacaacgtg	63
		Right:	cagcaaaaccccttcagcacaa	
EF628374	Epithelial calcium channel	Left:	gagaggccagaaggattcttga	109
		Right:	cagattccacttcagccttacga	
AY919670	60S ribosomal protein L8	Left:	ggctaaggtggttttccgtga	35
		Right:	cttcagctgcaatgaacagctc	
EF628372	Glucose-6-phosphate dehydrogenase	Left:	tgagggaagcctggagaatct	35
		Right:	ggtggcgtcttctctttctcg	
-	Cytochrome P4501A (CYP1A)	Left:	tctaacggttgtcccgatcct	9
		Right:	gaggcgcattagcagatacaca	
AY533140	Insulin-like growth factor-I	Left:	ggcaaaactccacgatcccta	9
		Right:	atgtccagatataggtttctttgctg	
-	microglobulin	Left:	aactgctgaaagatggagtggtt	3
		Right:	ccctttttcgaaggccaggt	
AY751300	Mx protein	Left:	gaaatggcatgggagaatcag	3
		Right:	cctgggcttcacgaatctttt	
AF130354	Vitellogenin precursor (Vtg)	Left:	ccatttgttcctgccactaagc	106
		Right:	cttgatgggaatctgaagctga	
AY643400	Elongation factor 1-alpha	Left:	ctctttctgttacctggcaaagg	66
		Right:	tcccatgattgattagtttcaggat	
AY855349	18S ribosomal RNA gene	Left:	cattggagggcaagtctggt	66
		Right:	tttaactgcagcaactttagtatacgc	
AY533141	Glucocorticoid receptor	Left:	caagaagggacgtcaaacagg	66
		Right:	ggaaagattgcgctctggaat	
-	Heat shock protein 90	Left:	ctggtcatcctcctgttcgag	56
		Right:	tgtgtctgaggatcgtccaatg	
-	Metallothionein	Left:	ctgccagtgtacaacatgcaaga	56
		Right:	gcacatttgctgcaaccaga	
-	Creatine kinase	Left:	ggaaatatgccacccccttcatt	5
		Right:	ccttgaacacctcataggtctcttc	
AF190773	Natural resistance associated macrophage protein	Left:	gcaatgcatcaggcagtcct	103
		Right:	ccacctgtagcgtgctgttgt	
AY643399	Growth hormone	Left:	gtggtcctggttagtttgttggt	103
		Right:	tgactgcgttgttgaagagcc	
AF192407	Zona pellucida glycoprotein 3 (ZP3)	Left:	atcatgggtgctttgtggatg	11
		Right:	gcttgtgaccgaggcatga	
EF628373	Glutathione S-transferase	Left:	ccggcaagagcttcaccat	48
		Right:	agtgaagtcgtgggaaataggc	
-	Aspartoacylase	Left:	tctggtaatggatgtcccgatt	94
		Right:	gacctctatggaaaagccatgc	
-	TGF beta	Left:	tgagcctgtacaacacgctga	1
		Right:	gtcccacgtagtagaggatggtg	

Statistical analysis: We used the geNorm algorithm to estimate the variability of reference gene expression, and to determine an optimal gene for normalization of the data (Vandesompele et al., 2002). Quantitative PCR data was analyzed using the relative quantification 2(-Delta Delta CT) method (Livak and Schmittgen, 2001). Expression was calculated relative to EF1alpha determined by GeNorm as the least variable gene in this study. As some quantitative PCR data were not distributed normally, significant differences in gene expression, relative to the unexposed controls, were assessed using Kruskal-Wallis nonparametric test with Dunn´s post-hoc test to compare treatments. For analysis each gene was treated as a separate experiment. Transcription profile heatmaps were created using Genesis gene expression analysis software (Technical University Graz, Austria).

Results and Discussion

Water chemistry: Physicochemical parameters measured at the beginning and end of the 24 h exposure period were within the acceptable range for the test organism (USEPA, 2002a; USEPA, 2002b) for all experiments and treatments. The measured mean values (± standard deviation) were pH: 7.51 (± 0.19), dissolved oxygen 7. 2 (± 0.5) $mg.L^{-1}$, temperature: 23.1 (± 0.3) ˚C, and EC: 278 (± 6) $\mu S.cm^{-1}$.

Sublethal effects: Expression of target genes involved in stress responses, neuromuscular function, and endocrine function, was determined, and insecticide-exposed fish were compared with untreated (control) fish. Individual effects were observed for each substance at concentrations below 50% of the LC_{10}. Concentration levels in the following sections refer to the measured dissolved fractions of active ingredient (A.I.), or to percentages of the nominal LC_{10} values determined by initial acute toxicity tests (table 2).

Table 2: Nominal and measured effect concentrations in [µg/L] of bifenthrin and fipronil for larval fathead minnow.

Substance	concentration [µg/L]	10% LC_{10}	20% LC_{10}	33% LC_{10}	50% LC_{10}
bifenthrin	nominal	0.09	0.18	0.31	0.46
	measured	0.07	0.14	0.24	0.35
fipronil	nominal	31	61	102	153
	measured	53	142	333	365

Effects on gene expression: Significant gene expression responses to bifenthrin and fipronil exposures were observed at the lowest tested concentrations, equal to 10% of the 24 h LC_{10}. The response was often bi-phasic, and genes that were down-regulated at the lowest exposure concentration were generally up-regulated at higher concentrations and vice versa. Beyond a threshold concentration, expression of most genes increased or decreased in a dose dependent manner. This threshold concentration was the same concentration that caused measurable negative effects on swimming ability of the fish (figures 1, 2; Beggel et al. 2010). In a few cases, expression levels remained unchanged at higher exposure concentrations after initial up- or down-regulation.

Gene responses to bifenthrin: Significant gene responses to bifenthrin exposure at only the lowest test concentration of 0.07 $\mu g.L^{-1}$ (10% LC_{10}) were measured for genes involved in endocrine function and growth: Cyp3A ($p<0.05$), parvalbumin ($p<0.01$), zona pellucida ($p<0.01$), glucocorticoid receptor ($p< 0.001$), insuline-like growth factor ($p<0.01$), growth hormone ($p<0.01$) and HSP90 ($p<0.001$). Expression of insuline-like growth factor ($p <0.001$) and growth hormone ($p <0.01$) was also significantly different from control at the highest exposure concentration.

Significant dose-dependent responses to bifenthrin were observed for genes involved in neuromuscular function, nerve repair and energetic status. Aspartoacylase (ASPA), in particular, was significantly downregulated at all exposure concentrations (0.07 $\mu g.L^{-1}$, $p <0.001$. 0.14 $\mu g.L^{-1}$, $p <0.01$, 0.24 $\mu g.L^{-1}$, $p <0.05$, 0.35 $\mu g.L^{-1}$, $p <0.05$). Aspartoacylase catalyzes hydrolysis of N-acetyl-L-aspartate (NAA) to aspartate and acetate in the vertebrate brain. Deficiency in ASPA activity leads to degeneration of Myelin sheet of nerve cell axons. Our data corroborate findings by Connon et.al. (2009), who observed significant downregulation of ASPA in the endangered fish species, delta smelt (*Hypomesus transpacificus*), after exposure to sublethal concentrations of the pyrethroid, esfenvalerate.

Creatine kinase was significantly downregulated after exposure to 0.24 $\mu g.L^{-1}$ ($p <0.05$) and 0.35 $\mu g.L^{-1}$ ($p <0.001$) bifenthrin. Creatine kinase plays an important role in the cellular energy metabolism, and is involved in muscle contraction by regulating calcium uptake and ATP/ADP ratios (Rossi et al., 1990). Creatine kinase activity is measured in humans to evaluate the energetic status of the brain. Expression of metallothionein was significantly up-regulated in fish exposed to 0.24 $\mu g.L^{-1}$ ($p <0.05$) and 0.35 $\mu g.L^{-1}$ ($p <0.001$) bifenthrin. Metallothionein is often used as a biomarker for metal-induced stress, but it is also known to contribute to a variety of other cellular processes, including neuroprotection and regeneration after CNS injuries (West et.al.2008).

Cytochrome CYP3A (Christen et al., 2010) plays an important role in the detoxification of xenobiotics in fish. Bifenthrin exposure led to a significant down-regulation of this enzyme at concentrations of 0.24 $\mu g.L^{-1}$ ($p<0.001$) and 0.35 $\mu g.L^{-1}$ ($p<0.001$). Additionally, another enzyme important in the cellular defense against oxidative stress, glutathione-S-transferase, was significantly down-regulated at the highest exposure concentration ($p <0.05$). Downregulation of such important enzymes has the potential to negatively affects the organism's detoxification capacity, and response to other stressors thus possible synergistic toxicity of bifenthrin with other chemicals is of concern.

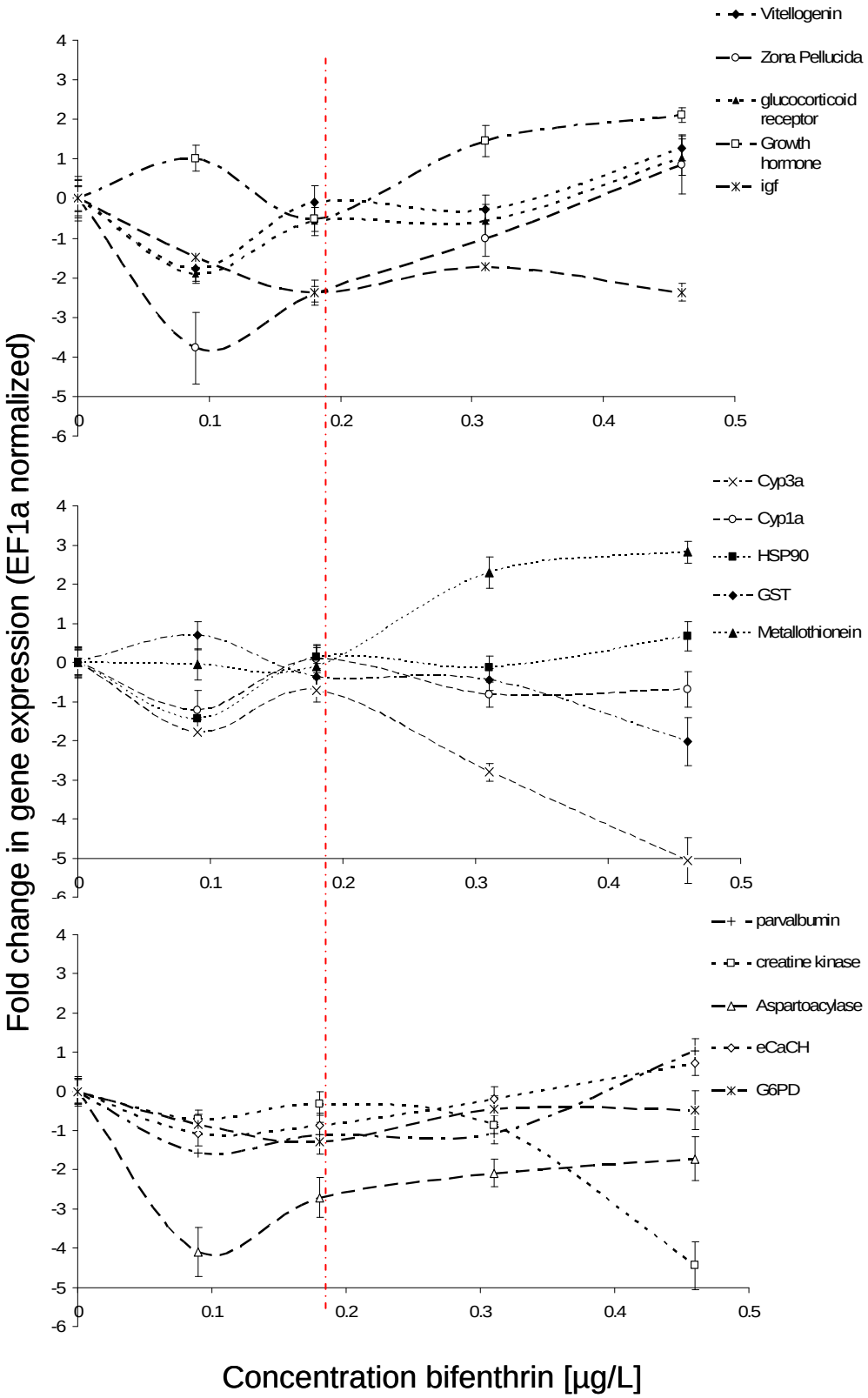

Figure 1: Changes in gene expression in reponse to bifenthrin exposure. Significant changes occurred at 0.07 µg.L^{-1}bifenthrin. Measured concentration levels ≥ 0.14 µg.L^{-1}result in negative effects on swimming performance (lowest observed effect threshold for swimming indicated by --). Strongest differences were observed for genes involved in detoxification, energy and neuromuscular function.

Gene responses to fipronil: Changes in gene expression occurred at the lowest test concentration of 53 $\mu g.L^{-1}$ fipronil (10% LC_{10}, measured concentration). Significant changes were observed for the epithelial calcium channel ($p < 0.001$), G6PD ($p < 0.05$), vitellogenin ($p < 0.05$) and metallothionein ($p < 0.05$).

At the highest fipronil concentration (365 $\mu g.L^{-1}$ measured concentration), significant up-regulation was observed for Cyp3A ($p < 0.01$), metallothionein ($p < 0.01$) and epithelial calcium channel ($p < 0.01$), suggesting that cellular xenobiotic detoxification processes and maintenance of ion homeostasis were negatively affected. Similar to bifenthrin-exposed fish, fipronil led to an up-regulation of ASPA at the highest exposure concentration ($p < 0.01$). Fipronil exposure also caused induction of the glucocorticoid receptor ($p < 0.001$); these are the principal receptors mediating the effects of glucocorticoids, mainly cortisol, in fish. Cortisol levels are used as indication of physiological stress, however, cortisol is also involved in growth, larval development, cognition and immune function. Glucocorticoids are known for maintaining basal and stress-related homeostasis in mammals. In the stressed state, elevated cortisol is important for central nervous system activation, increasing blood glucose concentration and elevating mean blood pressure, important parameters for stress compensation (Mommson et al., 1999). In fish, cortisol is involved in increasing plasma glucose concentration leading to increases in metabolic rate and oxygen uptake in response to elevated energy demand.
Exposure to fipronil also resulted in strong dose-dependent up-regulation of the gene coding for egg-yolk protein, or vitellogenin ($p < 0.01$), indicative of estrogenic effects in larval fish. Larval fish do not naturally express this gene.

From gene expression to higher level effects: Observed responses in gene expression demonstrate stress-related cellular effects in exposed fathead minnow larvae. The initial response at very low concentrations likely indicates disturbance in cell homeostasis leading, with increasing concentration, to adaptive and compensatory responses. It might be expected that these energy demanding responses result in measurable negative outcomes on the individual level, i.e. behavior, growth and reproduction. We recently demonstrated (Beggel et al. 2010) significant impacts of sublethal bifenthrin and fipronil concentrations on swimming ability and behavior, as well as 7-d growth of larval fathead minnow (Beggel et al. 2010). Swimming ability was significantly affected following 24 h exposure to ≥ 0.14 $\mu g.L^{-1}$ bifenthrin. This threshold corresponds to the onset of the second phase responses at the molecular level, which tended to be dose-dependent. Further assessment of swimming performance after 48 h and 6 d in control water showed that fish are able to recover from initial negative effects. The neuromuscular impairment after short bifenthrin exposure was reversible. This recovery was reflected in expression levels of ASPA. After the 6 d recovery period, the initially significantly downregulated ASPA showed no difference to control after exposure to 0.07 and 0.14 $\mu g.L^{-1}$ bifenthrin (figure 3).

Fipronil exposure led to significant impairment of swimming at 20% of the LC_{10}, however effects were less pronounced than in fish exposed to bifenthrin. Contrary to bifenthrin, recovery of swimming performance was not observed for fish exposed to 365 $\mu g.L^{-1}$ fipronil (measured). Fipronil exposed fish showed enhanced 7-d growth compared to controls, which may be linked to the strong estrogenic effects observed in the highest exposure concentration (figure 4). Expression of vitellogenin was increased approximately 90 fold after the 6 d recovery period .

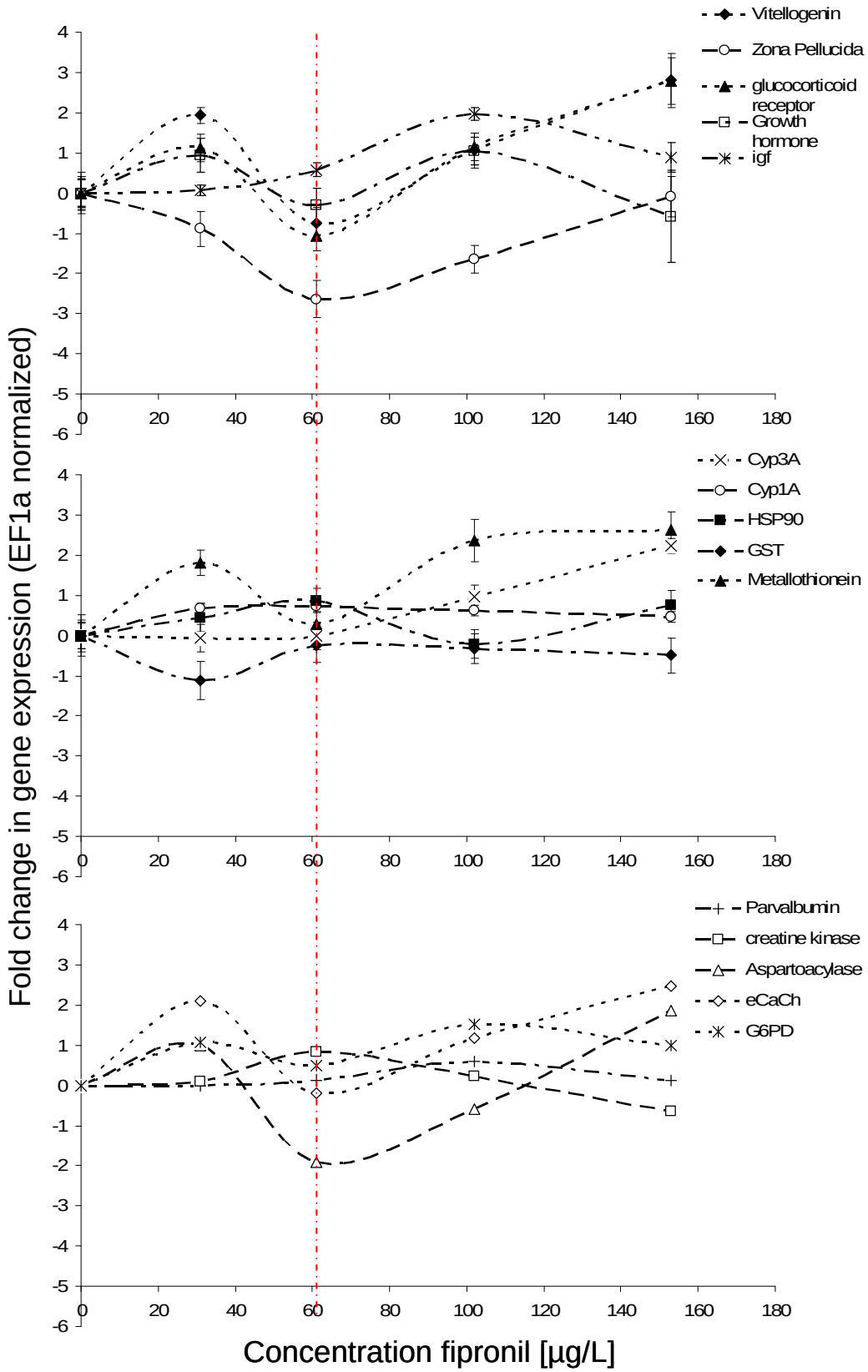

Figure 2: Changes in gene expression in reponse to fipronil exposure. Concentration levels ≥ 61 µg/L (nominal) resulted in negative effects on swimming performance (--). Strongest differences were observed for genes involved in detoxification and endocrine function.

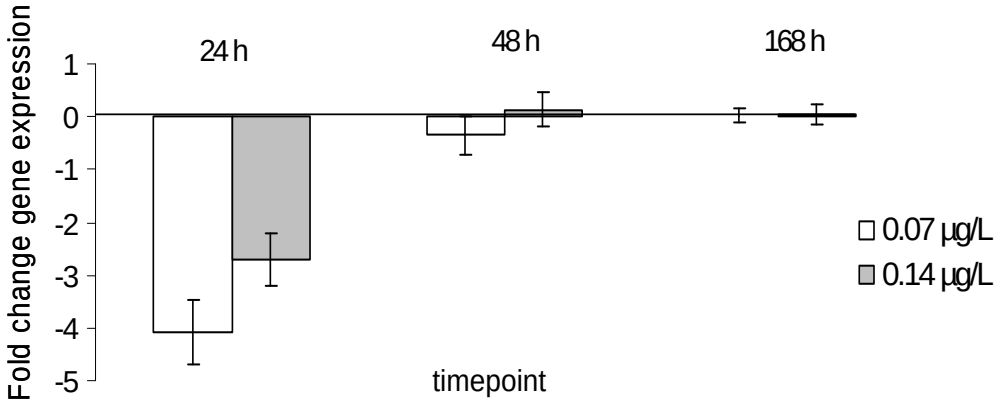

Figure 3: Fold change in gene expression of aspartoacylase exposed to 0.07 and 0.14 µg.L^{-1} bifenthrin. at three timepoints. (i) 24 h (exposure period); (ii) 48 h (24 h exposure, 24 h recovery); (iii) 168 h (24 h exposure, 6 d recovery).

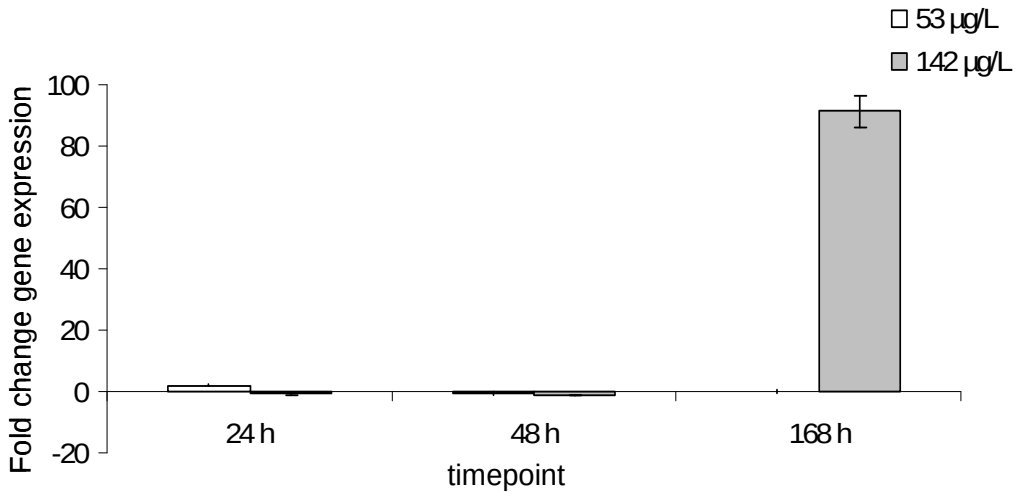

Figure 4: Fold change in gene expression of vitellogenin exposused to nominal 53 and 142 µg.L^{-1} fipronil at three timepoints. (i) 24 h (exposure period); (ii) 48 h (24 h exposure, 24 h recovery); (iii) 168 h (24 h exposure, 6 d recovery).

Expression profiling: Genesis software, a tool developed to analyze microarray datasets, was used to perform cluster analysis of gene expression data. Expression data from individual fish samples were grouped by hierarchical clustering to create a heatmap. This allowed discrimination between fish exposed to different insecticides. Two main clusters separated between bifenthrin and fipronil exposed fish (figure 5) with few exceptions. Within these two major groups, cluster analysis also distinguished between exposure concentrations. Due to individual variation in gene expression the transcriptional patterns were not exactly the same for all test animals from one specific treatment group, but the separation into distinct groups is obvious. However, further research is required to improve this approach. The application of this set of genes on field samples with unknown exposure history could be a promising method to screen for toxicant effects in the environment. Furthermore, the analysis of gene expression profiles for effects of chemical mixtures should be examined in more detail.

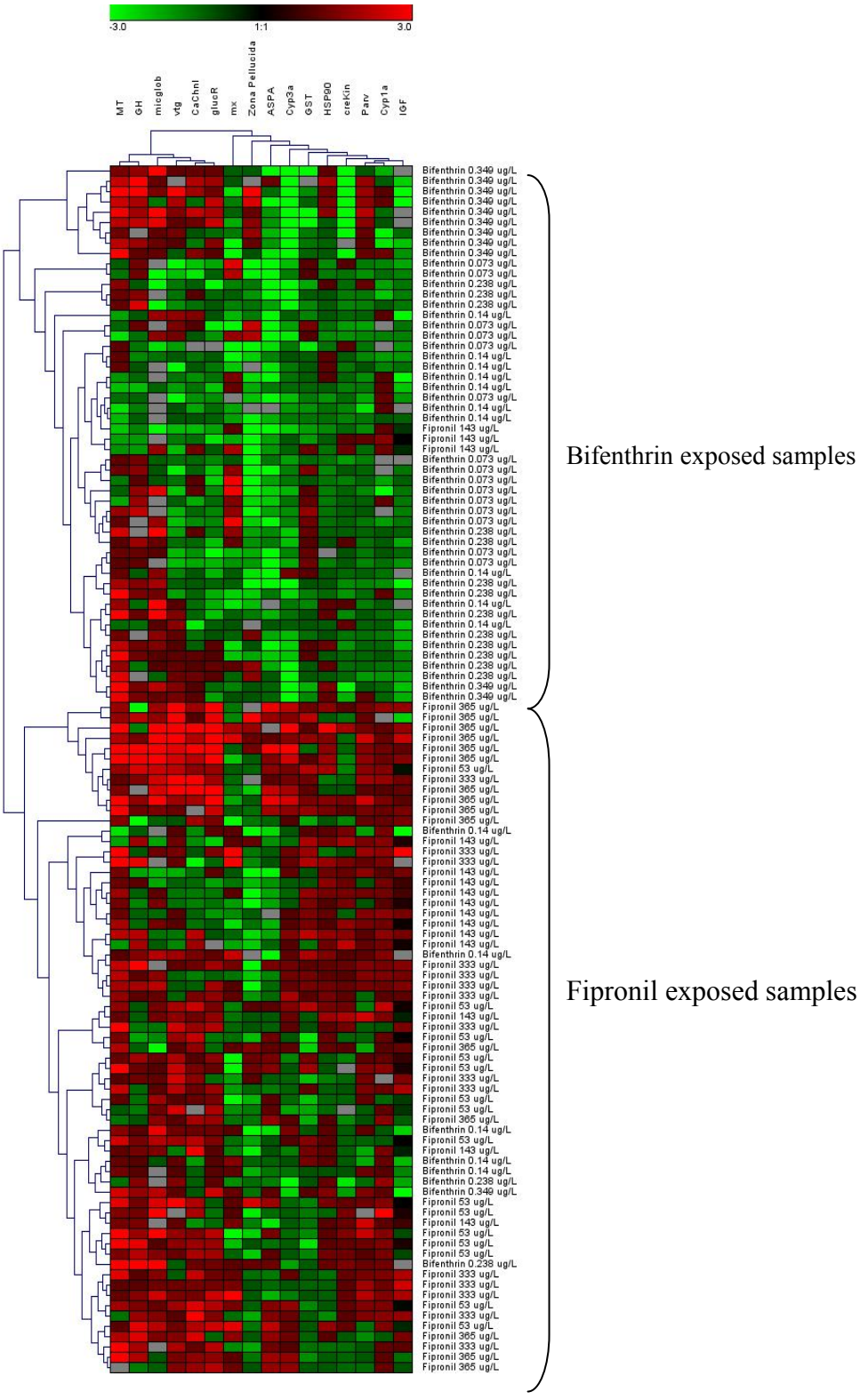

Figure 5: Heat map showing clusters of gene expression data from fish exposed to bifenthrin and fipronil (Manhattan distance) for 24 h. Red indicates up-regulation of genes, green down-regulation.

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DATA APPENDIX

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

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Appendix A

Analytical Chemistry and Laboratory Control Water Types

Table A-1. List of organophosphate pesticide analytes with corresponding method detection and reporting limits.

Organophosphate Pesticides	Method Detection Limit (µg/L)	Reporting limit (µg/L)
Azinphos methyl	0.030	0.050
Chlorpyrifos	0.010	0.020
Diazinon	0.005	0.020
Dimethoate	0.030	0.050
Disulfoton	0.010	0.050
Malathion	0.030	0.050
Methidathion	0.030	0.050
Parathion, Methyl	0.010	0.050
Phorate	0.030	0.050
Phosmet	0.030	0.050

Table A-2. List of pyrethroid pesticide analytes with corresponding method detection and reporting limits.

Pyrethroid Pesticides	Method Detection Limit (µg/L)	Reporting limit (µg/L)
Bifenthrin	0.001	0.002
Cyfluthrin	0.002	0.004
Cypermethrin	0.002	0.004
Deltamethrin	0.002	0.004
Esfenvalerate/Fenvalerate	0.001	0.002
Fenpropathrin	0.002	0.004
Lambda Cyhalothrin	0.001	0.002
Permethrin, Cis	0.003	0.005
Permethrin, Trans	0.003	0.005

Table A-3. List of carbamate pesticide analytes with corresponding method detection and reporting limits.

Carbamate Pesticides	Method Detection Limit (µg/L)	Reporting limit (µg/L)
Aldicarb	0.002	0.005
Captan	0.002	0.005
Carbaryl	0.001	0.002
Carbofuran	0.0005	0.001
Diuron	0.002	0.005
Linuron	0.002	0.005
Methiocarb	0.002	0.005
Methomyl	0.0005	0.001

Table A-4. List of Fipronil and Metabolites analytes with corresponding method detection and reporting limits.

Fipronil & Metabolites	Method Detection Limit (µg/L)	Reporting limit (µg/L)
Fipronil	0.100	0.200
Fipronil Desulfinyl	0.100	0.200
Fipronil Sulfide	0.100	0.200
Fipronil Sulfone	0.100	0.200

Table A-5. List of Trace Metal analytes with corresponding method detection and reporting limits.

Trace Metals	Method Detection Limit (µg/L)	Reporting limit (µg/L)
Aluminum	1.70	5.00
Arsenic	0.01	0.03
Cadmium	0.004	0.01
Chromium	0.10	0.30
Copper	0.03	0.10
Lead	0.002	0.006
Manganese	0.01	0.03
Nickel	0.01	0.03
Selenium	0.45	1.00
Silver	0.001	0.003
Zinc	0.05	0.15

Table A-6. List of PAH analytes with corresponding method detection and reporting limits.

PAHs	Method Detection Limit (µg/L)	Reporting limit (µg/L)
Naphthalene	0.00474	0.005
Methylnaphthalene, 2-	0.00457	0.005
Methylnaphthalene, 1-	0.00437	0.005
Dimethylnaphthalene, 2,6-	0.00293	0.005
Trimethylnaphthalene, 2,3,5-	0.00726	0.010
Naphthalenes, C1-	-	0.005
Naphthalenes, C2-	-	0.005
Naphthalenes, C3-	-	0.005
Naphthalenes, C4-	-	0.005
Biphenyl	0.00293	0.005
Acenaphthylene	0.00456	0.005
Acenaphthene	0.00251	0.005
Fluorene	0.00372	0.005
Methylfluorene, 1-	0.00656	0.010
Fluorenes, C1-	-	0.005
Fluorenes, C2-	-	0.005
Fluorenes, C3-	-	0.005
Dibenzothiophene	0.00195	0.005
Methyldibenzothiophene, 4-	0.00371	0.005
Dibenzothiophenes, C1-	-	0.005
Dibenzothiophenes, C2-	-	0.005
Dibenzothiophenes, C3-	-	0.005
Phenanthrene	0.00317	0.005
Methylphenanthrene, 1-	0.00762	0.010
Dimethylphenanthrene, 3,6-	0.00552	0.005
Phenanthrene/Anthracene, C1-	-	0.005
Phenanthrene/Anthracene, C2-	-	0.005
Phenanthrene/Anthracene, C3-	-	0.005
Phenanthrene/Anthracene, C4-	-	0.005
Anthracene	0.00281	0.005
Fluoranthene	0.00340	0.005
Methylfluoranthene, 2-	0.00410	0.005
Fluoranthene/Pyrenes, C1-	-	0.005
Pyrene	0.00379	0.005
Benz(a)anthracene	0.00364	0.005
Chrysene	0.00259	0.005
Chrysenes, C1-	-	0.005
Chrysenes, C2-	-	0.005
Chrysenes, C3-	-	0.005
Benzo(b)fluoranthene	0.00380	0.005
Benzo(k)fluoranthene	0.00377	0.005
Benzo(e)pyrene	0.00285	0.005
Benzo(a)pyrene	0.00345	0.005
Perylene	0.00313	0.005
Indenol(1,2,3-c,d)pyrene	0.00950	0.010
Dibenz(a,h)anthracene	0.00498	0.005
Benzo(g,h,i)perylene	0.00276	0.005

Table A-7. Description of synthetic laboratory control waters used for sensitivity studies.

Water	Species	SC-adjustment	pH-adjustment	Base
SSEPAMH	*C. dubia*	900 µS/cm with Instant Ocean	7.9 with hydrochloric acid or sodium hydroxide	Sierra Springs™ Spring Water adjusted to USEPA moderately hard standards
	E. affinis	900 µS/cm with Instant Ocean	7.9 with hydrochloric acid or sodium hydroxide	
SDEPAMH	*C. dubia*	900 µS/cm with Instant Ocean	7.9 with hydrochloric acid or sodium hydroxide	Sierra Springs™ Drinking Water adjusted to USEPA moderately hard standards
DIEPAMH	*P. promelas*	900 µS/cm with Instant Ocean	7.9 with hydrochloric acid or sodium hydroxide	Deionized water adjusted to USEPA moderately hard standards
DIEPAMHR	*H. azteca*	900 µS/cm with Instant Ocean	7.9 with hydrochloric acid or sodium hydroxide	Deionized water adjusted to USEPA moderately hard reconstituted standards
Filtered Hatchery Water	*H. transpacificus*	900 µS/cm with Instant Ocean	7.9 with hydrochloric acid or sodium hydroxide	Ambient water collected from the UC Davis Smelt Hatchery and passed through a 1µm filter.

Appendix B

Summary Tables:

Ambient Toxicity Tests with *H. Azteca*

Table B 1-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 1/03/08 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 1/01/08 - 1/02/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	90	0.5	89	4.5	NS
DIEPAMHR + organic matter	98	2.5	95	3.1	NS
High EC Control @ 13.15 mS/cm	97	2.8	100	0.0	NS
Sacramento River at Hood DWR Station	90	7.1	97	2.8	NS
Suisun Bay at Public Suisun Dock	97	3.1	100	0.0	NS
Napa River, near River Park Blvd.[3]	100	0.0	95	5.0	NS
Sacramento River at tip of Grand Island (711)	98	2.5	100	0.0	NS
San Joaquin River between Hog and Turner Cuts (910)	97	2.8	95	3.1	NS
Confluence of Linsey Sl. And Cache Sl.	85	4.9	93	4.8	NS
San Joaquin River at Potato Slough (815)	100	0.0	100	0.0	NS
Sacramento Deep Water Channel, Light 55	97	2.8	90	5.8	NS
Old River at mouth of Holland Cut (915)	100	0.0	98	2.5	NS
Old River, western arm at railroad bridge (902)	98	2.5	100	0.0	NS
Upper Cache Slough at mouth of Ulatis Creek	95	2.9	100	0.0	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.032	0.015	0.047	0.009	NS
DIEPAMHR + organic matter	0.056	0.007	0.055	0.007	NS
High EC Control @ 13.15 mS/cm	0.052	0.013	0.051	0.009	NS
Sacramento River at Hood DWR Station	0.034	0.009	0.062	0.006	S*
Suisun Bay at Public Suisun Dock	0.045	0.014	0.057	0.009	NS
Napa River, near River Park Blvd.[3]	0.062	0.009	0.069	0.011	NS
Sacramento River at tip of Grand Island (711)	0.078	0.008	0.087	0.004	NS
San Joaquin River between Hog and Turner Cuts (910)	0.071	0.010	0.088	0.002	NS
Confluence of Linsey Sl. And Cache Sl.	0.067	0.005	0.074	0.007	NS
San Joaquin River at Potato Slough (815)	0.087	0.009	0.091	0.005	NS
Sacramento Deep Water Channel, Light 55	0.072	0.007	0.079	0.003	NS
Old River at mouth of Holland Cut (915)	0.081	0.005	0.100	0.004	S*
Old River, western arm at railroad bridge (902)	0.071	0.011	0.087	0.008	NS
Upper Cache Slough at mouth of Ulatis Creek	0.070	0.012	0.096	0.009	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.
3. This high conductivity sample was compared to the high EC control.

Table B 1-2. Summary of 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 1/01/08 - 1/02/08.

Treatment	Collection Date & Time	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 Hood DWR Station	1/01/08 11:10	212	8.8	7.37	11.7	14.3	0.51	0.002
Suisun Bay at Public Suisun Dock	1/01/08 17:30	5960	7.3	7.65	10.2	266.3	0.32	0.002
Napa River, near River Park Blvd.	1/01/08 16:20	13470	9.2	7.17	10.5	69.5	0.37	0.001
Sacramento River at tip of Grand Island (711)	1/02/08 12:38	386	7.5	7.40	12.3	11.8	0.41	0.001
San Joaquin River between Hog and Turner Cuts (910)	1/02/08 10:30	610	7.4	7.55	12.5	5.7	0.13	0.001
Confluence of Linsey Sl. And Cache Sl.	1/02/08 13:20	379	7.7	7.76	12.6	16.1	0.44	0.004
San Joaquin River at Potato Slough (815)	1/02/08 11:35	722	7.9	7.72	12.3	7.7	0.23	0.002
Sacramento Deep Water Channel, Light 55	1/02/08 13:00	387	7.9	7.75	12.5	18.6	0.44	0.004
Old River at mouth of Holland Cut (915)	1/02/08 9:45	888	6.9	7.44	12.4	5.6	0.14	0.001
Old River, western arm at railroad bridge (902)	1/02/08 9:15	1444	6.9	7.56	12.4	6.1	0.19	0.001
Upper Cache Slough at mouth of Ulatis Creek	1/02/08 13:45	478	8.7	7.95	12.7	8.1	0.13	0.002

Table B 1-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 1/03/08 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 1/01/08 - 1/02/08.

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	22.2	22.3	7.6	8.9	7.78	8.15	104	56	-
DIEPAMHR + organic matter	343	22.6	22.9	7.6	8.9	7.71	8.14	104	56	-
High EC Control @ 13.15 mS/cm	12520	22.6	23.5	7.5	8.7	7.73	8.02	1520	72	-
Sacramento River at Hood DWR Station	314	22.3	22.6	7.5	8.8	7.86	8.11	76	82	0.016
Suisun Bay at Public Suisun Dock	5640	22.6	22.8	7.6	8.6	7.99	8.34	768	208	0.011
Napa River, near River Park Blvd.	12040	22.1	22.6	7.3	8.7	7.81	7.98	1660	112	0.009
Sacramento River at tip of Grand Island (711)	294	22.1	22.5	7.5	8.9	7.88	8.12	88	82	0.015
San Joaquin River between Hog and Turner Cuts (910)	500	22.2	23.3	7.4	8.8	7.91	8.12	120	88	0.006
Confluence of Linsey Sl. And Cache Sl.	244	22.3	23.3	7.3	8.8	7.84	8.16	84	88	0.028
San Joaquin River at Potato Slough (815)	592	22.4	22.6	7.6	8.8	7.84	8.08	116	78	0.011
Sacramento Deep Water Channel, Light 55	242	22.3	22.4	7.5	8.9	7.88	8.17	92	90	0.019
Old River at mouth of Holland Cut (915)	767	22.2	22.9	7.7	8.8	7.83	8.10	132	76	0.007
Old River, western arm at railroad bridge (902)	875	22.3	23.4	7.7	8.7	7.83	8.05	144	80	0.007
Upper Cache Slough at mouth of Ulatis Creek	400	22.4	22.7	7.5	8.8	8.09	8.27	128	118	0.008
DIEPAMHR + 25 ppb PBO	332	22.3	22.4	7.6	8.9	7.77	8.12	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	332	22.4	22.5	7.6	8.8	7.74	8.12	-	-	-
High EC Control @ 13.15 mS/cm + 25 ppb PBO	11700	22.3	22.9	7.9	8.9	7.71	8.02	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	219	22.2	22.7	7.5	8.8	7.88	8.12	-	-	-
Suisun Bay at Public Suisun Dock + 25 ppb PBO	5345	22.1	22.5	7.4	8.7	8.02	8.33	-	-	-
Napa River, near River Park Blvd. + 25 ppb PBO	12345	22.1	22.7	7.2	8.9	7.82	7.93	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	304.9	22.0	22.5	7.5	8.7	7.82	8.11	-	-	-
San Joaquin River between Hog and Turner Cuts (910) + 25 ppb PBO	498	22.1	23.1	7.4	8.9	7.89	8.14	-	-	-
Confluence of Linsey Sl. And Cache Sl. + 25 ppb PBO	271	22.1	23.5	7.6	8.8	7.86	8.14	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	581	21.8	23.5	7.2	8.9	7.85	8.06	-	-	-
Sacramento Deep Water Channel, Light 55 + 25 ppb PBO	244	22.0	23.7	7.4	8.9	7.90	8.14	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	765	22.0	23.5	7.7	8.9	7.83	8.07	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	887	21.9	23.1	7.7	8.9	7.84	8.07	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	399	22.1	23.5	7.6	8.8	8.08	8.27	-	-	-

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 B 2-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 1/04/08 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 1/03/08 - 1/04/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	90	4.1	88	2.6	NS
DIEPAMHR + organic matter	95	2.9	95	2.8	NS
High EC Control @ 11.0 mS/cm	100	0.0	98	2.5	NS
High EC Control @ 22.0 mS/cm	92	2.6	81*	5.4	NS
Montezuma Slough at Nurse Slough (609)[3]	98	2.5	95	3.1	NS
Suisun Bay off Chipps Island (508)[3]	95	2.9	100	0.0	NS
Grizzly Bay at Dolphin (602)[4]	100	0.0	95	3.1	NS
Rough and Ready DWR station, Stockton	100	0.0	100	0.0	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.030	0.005	0.027**	0.006	NS
DIEPAMHR + organic matter	0.044	0.007	0.052	0.002	NS
High EC Control @ 11.0 mS/cm	0.043	0.007	0.045	0.006	NS
High EC Control @ 22.0 mS/cm	0.039	0.003	0.032*	0.006	NS
Montezuma Slough at Nurse Slough (609)[3]	0.046	0.007	0.048	0.001	NS
Suisun Bay off Chipps Island (508)[3]	0.063	0.003	0.054	0.004	NS
Grizzly Bay at Dolphin (602)[4]	0.046	0.008	0.039	0.006	NS
Rough and Ready DWR station, Stockton	0.053	0.004	0.063	0.006	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

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

3. These high conductivity samples were compared to the High EC Control @ 11.0 mS/cm.

4. This high conductivity sample was compared to the High EC Control @ 22.0 mS/cm.

Table B 2-2. Summary of 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 1/03/08 - 1/04/08.

Treatment	Collection Date & Time	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)	1/03/08 12:15	14390	9.1	7.35	11.6	27.2	0.27	0.005
Suisun Bay off Chipps Island (508)	1/03/08 9:45	10590	8.3	7.79	12.0	10.1	0.21	0.006
Grizzly Bay at Dolphin (602)	1/03/08 11:30	22140	9.0	7.91	11.7	17.5	0.17	0.004
Rough and Ready DWR station, Stockton	1/04/08 9:30	846	9.0	7.78	11.1	4.6	0.16	0.005

Table B 2-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 1/04/08 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 1/03/08 - 1/04/08.

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	333	22.3	22.5	7.6	8.8	7.79	8.07	104	56	-
DIEPAMHR + organic matter	334	22.3	22.7	7.5	8.9	7.68	8.02	104	56	-
High EC Control @ 11.0 mS/cm	10195	22.5	23.0	7.6	8.7	7.73	7.92	1200	72	-
High EC Control @ 22.0 mS/cm	20400	22.4	23.1	7.2	8.7	7.74	7.98	2400	84	-
Montezuma Slough at Nurse Slough (609)	10555	22.3	22.4	7.2	8.8	7.74	7.93	1220	92	0.005
Suisun Bay off Chipps Island (508)	9955	22.4	23.1	7.3	8.9	7.82	7.93	1120	90	0.006
Grizzly Bay at Dolphin (602)	19230	22.3	23.1	7.0	8.7	7.75	7.91	2400	98	0.004
Rough and Ready DWR station, Stockton	810	22.2	23.0	7.7	8.9	7.90	8.10	176	104	0.005
DIEPAMHR + 25 ppb PBO	339	22.2	22.5	7.6	9.5	7.82	8.03	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	336	22.1	22.2	7.4	8.8	7.70	8.08	-	-	-
High EC Control @ 11.0 mS/cm + 25 ppb PBO	10095	22.2	22.2	7.2	8.8	7.70	7.95	-	-	-
High EC Control @ 22.0 mS/cm + 25 ppb PBO	20170	22.2	22.3	7.2	8.8	7.72	7.99	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	10060	22.1	22.7	7.4	8.9	7.77	7.94	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	9680	22.1	22.4	7.3	8.8	7.82	7.95	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	19350	22.1	22.2	7.0	8.7	7.74	7.90	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	818	22.1	22.5	7.6	8.9	7.89	8.10	-	-	-

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 B 3-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 01/17/08 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 01/15/08 - 01/16/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	98	2.3	95	2.9	NS
DIEPAMHR + POM	98	2.5	90	0.0	NS
High EC Control @ 20.80 mS/cm + POM	97	2.8	100	0.0	NS
Suisun @ Public Access	98	2.5	100	0.0	NS
Napa @ Valley College	94	3.8	100	0.0	NS
Suisun Bay off Chipps Island (508)	100	0.0	98	2.5	NS
Grizzly Bay @ Dolphin (602)	98	2.5	95	2.9	NS
Carquinez Strait, West of Benecia army dock (405)[3]	97	2.8	100	0.0	NS
Napa River @ Vallejo Seawall (340)[3]	100	0.0	89	4.6	NS
Montezuma Slough at Nurse Slough (609)	100	0.0	100	0.0	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.055*	0.004	0.053*	0.004	NS
DIEPAMHR + POM	0.066	0.003	0.066	0.002	NS
High EC Control @ 20.80 mS/cm + POM	0.038***	0.003	0.038***	0.003	NS
Suisun @ Public Access	0.088	0.006	0.104	0.004	S*
Napa @ Valley College	0.100	0.003	0.107	0.007	NS
Suisun Bay off Chipps Island (508)	0.098	0.004	0.094	0.004	NS
Grizzly Bay @ Dolphin (602)	0.096	0.007	0.085	0.001	NS
Carquinez Strait, West of Benecia army dock (405)[3]	0.063	0.010	0.052	0.006	NS
Napa River @ Vallejo Seawall (340)[3]	0.064	0.003	0.065	0.007	NS
Montezuma Slough at Nurse Slough (609)	0.079	0.010	0.066	0.010	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

***: $P < 0.001$

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

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

Table B 3-2. Summary of 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 01/15/08-01/16/08.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Suisun @ Public Access	2887	10.5	7.3	8.4	56.3	0.30	0.002
Napa @ Valley College	590	11.6	7.45	10.4	90.9	0.36	0.001
Suisun Bay off Chipps Island (508)	1045	7.8	7.32	11.1	82.3	0.27	0.001
Grizzly Bay @ Dolphin (602)	4023	8.9	7.54	11.1	35.9	0.08	0.000
Carquinez Strait, West of Benecia army dock (405)	20190	9.0	7.57	10.4	11.9	0.31	0.001
Napa River @ Vallejo Seawall (340)	21740	9.1	7.62	10.3	58.4	0.27	0.001
Montezuma Slough at Nurse Slough (609)	7710	8.7	7.27	10.3	45.7	0.25	0.001

Table B 3-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 01/17/08 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 01/15/08-01/16/08.

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	249	22.2	23.4	7.6	8.4	7.22	8.10	104	56	-
DIEPAMHR + 1% nutrient add back	250	22.3	22.8	7.5	8.5	7.21	8.02	104	56	-
High EC Control @ 20.80 mS + 1% nutrient add back	18720	20.5	22.3	6.8	8.7	7.13	7.95	2240	84	-
Suisun @ Public Access	2682	22.3	22.7	7.5	8.7	7.44	8.33	388	166	0.005
Napa @ Valley College	535	22.2	23.0	7.6	8.8	7.32	8.07	132	78	0.003
Suisun Bay off Chipps Island (508)	806	20.2	22.1	7.8	8.9	7.25	8.04	160	74	0.010
Grizzly Bay @ Dolphin (602)	4361	22.0	23.4	7.6	8.7	7.27	8.04	528	84	0.008
Carquinez Strait, West of Benecia army dock (405)	17810	22.0	23.5	7.2	8.2	7.23	7.93	2040	96	0.005
Napa River @ Vallejo Seawall (340)	19450	22.0	23.7	6.9	8.6	7.24	7.91	2320	98	0.007
Montezuma Slough at Nurse Slough (609)	7115	22.0	23.4	7.4	8.4	7.19	8.03	860	82	0.014
DIEPAMHR + 25 ppb PBO	347	21.9	23.8	7.7	8.4	7.21	8.03	-	-	-

DIEPAMHR + 1% nutrient add back + 25 ppb PBO	336	21.9	23.8	7.6	8.8	7.25	8.07	-	-	-
High EC Control @ 20.80 mS + 1% nutrient add back + 25 ppb PBO	19180	21.8	23.3	7.3	8.4	7.16	7.99	-	-	-
Suisun @ Public Access + 25 ppb PBO	7353	19.8	21.8	7.6	8.8	7.55	8.36	-	-	-
Napa @ Valley College + 25 ppb PBO	551	21.7	23.0	7.5	8.7	7.33	8.06	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	1102	21.7	24.0	7.6	8.6	7.26	8.08	-	-	-
Grizzly Bay @ Dolphin (602) + 25 ppb PBO	4270	21.7	23.0	7.7	8.6	7.25	8.03	-	-	-
Carquinez Strait, West of Benecia army dock (405)3 + 25 ppb PBO	17815	21.6	23.8	7.4	8.7	7.22	7.95	-	-	-
Napa River @ Vallejo Seawall (340)3 + 25 ppb PBO	19340	21.6	23.8	7.0	8.7	7.23	7.90	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	7025	21.5	22.3	7.4	8.6	7.17	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 B 4-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 01/18/08 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 01/17/08 - 01/18/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	95	4.5	95	2.9	NS
DIEPAMHR + POM	93	4.8	95	2.9	NS
San Joaquin River @ Potato Slough (815)	98	2.5	95	2.9	NS
Old River, western arm at railroad bridge (902)	100	0.0	98	2.5	NS
Confluence of Lindsey slough and Cache slough	95	2.9	93	2.5	NS
Old River at mouth of Holland Cut (915)	100	0.0	100	0.0	NS
Upper Cache Slough at mouth of Ulatis Creek (CU)	98	2.3	95	2.8	NS
Sacramento River, Deep Water Channel, Light 55	93	4.8	98	2.5	NS
Sacramento River at tip of Grand Island (711)	95	5.0	95	2.9	NS
Sacramento River at Hood DWR Station	98	2.5	93	2.5	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.036*	0.003	0.033**	0.006	NS
DIEPAMHR + POM	0.061	0.008	0.060	0.003	NS
San Joaquin River @ Potato Slough (815)	0.069	0.005	0.089	0.008	NS
Old River, western arm at railroad bridge (902)	0.074	0.005	0.079	0.011	NS
Confluence of Lindsey slough and Cache slough	0.060	0.004	0.064	0.009	NS
Old River at mouth of Holland Cut (915)	0.072	0.007	0.079	0.012	NS
Upper Cache Slough at mouth of Ulatis Creek (CU)	0.066	0.006	0.067	0.006	NS

Sacramento River, Deep Water Channel, Light 55	0.060	0.004	0.066	0.012	NS
Sacramento River at tip of Grand Island (711)	0.061	0.002	0.056	0.004	NS
Sacramento River at Hood DWR Station	0.084	0.017	0.085	0.013	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

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

Table B 4-2. Summary of 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 01/17/08 and 01/18/08.

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 at Potato Slough (815)	329	8.3	7.38	11.9	34.5	0.14	0.001
Old River, western arm at railroad bridge (902)	500	8.1	7.50	11.9	31.9	0.13	0.001
Confluence of Lindsey Sl. And Cache Sl.	255	8.4	7.52	12.1	64.4	0.24	0.001
Old River at mouth of Holland Cut (915)	538	8.2	7.48	11.7	14.6	0.44	0.002
Upper Cache Slough at mouth of Ulatis Creek	392	7.8	7.83	12.1	95.9	0.15	0.002
Sacramento Deep Water Channel, Light 55	303	8.4	7.64	11.8	90.0	0.18	0.001
Sacramento River at tip of Grand Island (711)	218	8.2	7.39	12.2	40.8	0.28	0.001
Sacramento River at Hood DWR Station	234	9.5	7.49	11.3	27.6	0.43	0.002

Table B 4-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 01/18/08 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 01/17/08 and 01/18/08.

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			
Control (DIEPAMHR)	331	21.9	22.4	7.7	8.6	7.82	8.08	104	56	-
Control (DIEPAMHR + Organic Matter)	336	22.1	22.4	7.7	8.6	7.84	8.04	104	56	-

San Joaquin River at Potato Slough (815)	314	22.2	22.8	7.5	8.7	7.89	7.98	88	68	0.006
Old River, western arm at railroad bridge (902)	476	22.1	22.8	7.7	8.8	7.91	8.03	100	76	0.005
Confluence of Lindsey Sl. And Cache Sl.	245	21.6	21.8	7.6	8.8	7.90	8.12	84	84	0.008
Old River at mouth of Holland Cut (915)	531	21.7	22.2	7.6	8.6	7.89	8.00	108	76	0.014
Upper Cache Slough at mouth of Ulatis Creek	433	21.4	22.6	7.6	8.7	8.06	8.36	136	120	0.007
Sacramento Deep Water Channel, Light 55	317	21.9	22.7	7.5	8.5	8.06	8.23	104	98	0.009
Sacramento River at tip of Grand Island (711)	210	21.9	22.7	7.5	8.9	7.87	8.18	76	98	0.009
Sacramento River at Hood DWR Station	230	21.8	22.8	7.5	8.7	7.85	8.12	84	86	0.013
Control (DIEPAMHR) + 25ppb PBO	346	21.7	21.8	7.9	8.9	7.84	8.11	-	-	-
Control (DIEPAMHR + Organic Matter) + 25ppb PBO	334	21.6	21.7	7.7	8.8	7.79	8.09	-	-	-
San Joaquin River at Potato Slough (815) + 25ppb PBO	316	21.6	22.3	7.5	8.7	7.89	8.06	-	-	-
Old River, western arm at railroad bridge (902) + 25ppb PBO	476	21.7	22.4	7.7	8.7	7.94	8.05	-	-	-
Confluence of Linsey Sl. And Cache Sl. + 25ppb PBO	249	21.7	22.4	7.6	8.7	7.97	8.22	-	-	-
Old River at mouth of Holland Cut (915) + 25ppb PBO	525	21.5	22.3	7.6	8.8	7.88	8.04	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25ppb PBO	431	21.3	22.4	7.4	8.7	8.09	8.33	-	-	-
Sacramento Deep Water Channel, Light 55 + 25ppb PBO	318	21.5	22.0	7.4	8.7	8.04	8.25	-	-	-
Sacramento River at tip of Grand Island (711) + 25ppb PBO	222	21.4	22.5	7.3	8.8	7.88	8.13	-	-	-
Sacramento River at Hood DWR Station + 25ppb PBO	227	21.5	23.0	7.3	8.7	7.72	8.16	-	-	-

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

Table B 5-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 01/31/08 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 01/29/08 - 01/30/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	95	2.9	93	4.8	NS
DIEPAMHR + POM	95	2.9	90	4.1	NS
High EC Control @ 14.28 mS/cm + POM	100	0.0	100	0.0	NS
Suisun @ Rush Ranch	98	2.3	98	2.3	NS
Montezuma Slough at Nurse Slough (609)	100	0.0	100	0.0	NS
Napa River at Vallejo Seawall (340)[3]	98	2.5	100	0.0	NS
Grizzly Bay at Dolphin (602)	100	0.0	100	0.0	NS
Suisun Bay off Chipps Island (508)	100	0.0	98	2.5	NS
Carquinez Strait, West of Benicia army dock (405)[3]	100	0.0	100	0.0	NS
Field Duplicate: Napa River at Vallejo Seawall (340)[3]	100	0.0	-	-	NA
Trip Blank (DIEPAMHR)	100	0.0	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.057*	0.007	0.062**	0.003	NS
DIEPAMHR + POM	0.078	0.005	0.093	0.005	NS
High EC Control @ 14.28 mS/cm + POM	0.079	0.006	0.073	0.009	NS
Suisun @ Rush Ranch	0.096	0.004	0.105	0.006	NS
Montezuma Slough at Nurse Slough (609)	0.099	0.005	0.081	0.006	NS
Napa River at Vallejo Seawall (340)[3]	0.078	0.005	0.087	0.002	NS
Grizzly Bay at Dolphin (602)	0.103	0.011	0.101	0.006	NS
Suisun Bay off Chipps Island (508)	0.097	0.006	0.107	0.006	NS
Carquinez Strait, West of Benicia army dock (405)[3]	0.079	0.007	0.080	0.003	NS
Field Duplicate: Napa River at Vallejo Seawall (340)[3]	0.089	0.005	-	-	NA
Trip Blank (DIEPAMHR)	0.061	0.004	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

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

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

Table B 5-2. Summary of 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 01/29/08-01/31/08.

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 at Public Suisun Dock (SU)	3516	8.3	7.34	10.1	69.5	0.12	0.000
Trip Blank (337)	342	16.1	8.06	10.1	-	0.01	0.000
Suisun Bay off Chipps Island (508)	678	8.0	7.40	12.1	38.7	0.29	0.001
Montezuma Slough at Nurse Slough (609)	5830	8.9	7.10	11.2	40.1	0.27	0.000
Grizzly Bay at Dolphin (602)	2723	8.3	6.95	12.3	57.3	0.29	0.000
Napa River at Vallejo Seawall (340)	11740	8.7	7.09	11.2	50.7	0.25	0.000
Field Duplicate: Napa River at Vallejo Seawall (340)	11740	8.7	7.09	11.2	61.9	0.26	0.000
Carquinez Strait, West of Benicia army dock (405)	13430	8.5	7.34	11.7	27.3	0.24	0.001

Table B 5-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 01/31/08 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 01/29/08-01/30/08.

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	22.2	24.1	7.8	8.8	6.94	8.13	108	72	-
DIEPAMHR + 1% organic matter	278	22.0	23.2	7.6	8.6	6.74	8.19	108	72	-
High EC Control @ 14.28 mS + 1% organic matter	12960	22.2	22.3	7.5	8.3	6.75	8.03	1520	74	-
Suisun @ Rush Ranch	3306	22.7	22.9	7.8	8.6	7.41	8.23	424	166	0.004
Montezuma Slough at Nurse Slough (609)	5350	22.9	23.8	7.5	8.7	7.76	7.89	640	80	0.006
Napa River at Vallejo Seawall (340)	10525	22.0	23.7	7.5	8.6	7.78	7.90	1400	82	0.006
Grizzly Bay at Dolphin (602)	2435	22.8	23.4	7.8	8.5	7.90	8.06	304	82	0.013
Suisun Bay off Chipps Island (508)	540	22.6	23.8	7.6	8.9	7.99	8.06	112	86	0.015
Carquinez Strait, West of Benicia army dock (405)	12440	22.6	23.7	7.5	8.4	7.86	7.92	1640	88	0.006
Field Duplicate: Napa River at Vallejo Seawall (340)	10460	22.0	23.6	7.6	8.5	7.78	7.91	1240	80	0.006
Trip Blank (DIEPAMHR)	371	22.0	23.8	8.0	8.5	7.90	8.21	108	60	0.001
DIEPAMHR + 25 ppb PBO	332	22.0	22.8	7.9	8.4	7.87	8.11	-	-	-
DIEPAMHR + 1% organic matter + 25 ppb PBO	340	22.6	22.8	7.7	8.4	7.81	8.09	-	-	-
High EC Control @ 14.28 mS + 1% organic matter + 25 ppb PBO	13070	22.5	22.8	7.6	8.3	7.74	8.00	-	-	-
Suisun @ Rush Ranch + 25 ppb PBO	3318	22.6	23.3	7.8	8.4	7.96	8.32	-	-	-

Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	5300	22.4	23.2	7.4	8.3	7.78	7.89	-	-	-
Napa River at Vallejo Seawall (340) + 25 ppb PBO	10585	22.6	23.2	7.6	8.3	7.78	7.88	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	2404	22.7	23.3	7.7	8.8	7.92	8.03	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	2815	22.4	23.1	7.6	8.6	8.03	8.23	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	12245	22.6	23.6	7.6	8.4	7.83	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 B 6-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 2/01/08 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 1/31/08 - 2/01/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	95	2.8	95	2.9	NS
DIEPAMHR + organic matter	100	0.0	98	2.5	NS
Old River at mouth of Holland Cut (915)	100	0.0	100	0.0	NS
San Joaquin River at Potato Slough (815)	100	0.0	100	0.0	NS
Old River, western arm at railroad bridge (902)	100	0.0	98	2.5	NS
Sacramento River at tip of Grand Island (711)	90	7.1	92	4.8	NS
Confluence of Linsey Sl. And Cache Sl.	98	2.5	95	2.9	NS
Upper Cache Slough at mouth of Ulatis Creek	97	2.8	8*	4.9	S (8%)*
Sacramento Deep Water Channel, Light 55	100	0.0	91	9.1	NS
Sacramento River at Hood DWR Station	100	0.0	90	4.1	NS
Rough and Ready DWR station, Stockton	100	0.0	100	0.0	NS
Napa River, near River Park Blvd.	100	0.0	98	2.5	NS
Bottle Blank: DIEPAMHR	95	2.9	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.060*	0.003	0.045**	0.010	NS
DIEPAMHR + organic matter	0.082	0.009	0.088	0.004	NS
Old River at mouth of Holland Cut (915)	0.091	0.005	0.091	0.004	NS
San Joaquin River at Potato Slough (815)	0.092	0.006	0.080	0.008	NS
Old River, western arm at railroad bridge (902)	0.093	0.011	0.085	0.011	NS
Sacramento River at tip of Grand Island (711)	0.067	0.010	0.065**	0.005	NS
Confluence of Linsey Sl. And Cache Sl.	0.088	0.010	0.067**	0.004	NS
Upper Cache Slough at mouth of Ulatis Creek	0.042**	0.007	0.055**	0.095	NS

Sacramento Deep Water Channel, Light 55	0.090	0.010	0.081	0.004	NS
Sacramento River at Hood DWR Station	0.083	0.004	0.081	0.006	NS
Rough and Ready DWR station, Stockton	0.084**	0.005	0.076*	0.004	NS
Napa River, near River Park Blvd.	0.075	0.011	0.063**	0.006	NS
Bottle Blank: DIEPAMHR	0.048	0.011	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

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

Table B 6-2. Summary of 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 1/31/08 - 2/01/08.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (^oC)	pH	DO (mg/L)			
Old River at mouth of Holland Cut (915)	429	8.3	6.85	12.1	17.1	0.14	0.000
San Joaquin River at Potato Slough (815)	263	8.1	6.75	11.9	80.2	0.21	0.000
Old River, western arm at railroad bridge (902)	399	7.9	7.01	12.2	18.4	0.12	0.000
Sacramento River at tip of Grand Island (711)	150	8.0	6.89	11.9	143.0	0.24	0.000
Confluence of Lindsey Sl. And Cache Sl.	263	7.9	7.00	11.8	217.3	0.24	0.000
Upper Cache Slough at mouth of Ulatis Creek	347	8.5	6.80	11.1	119.0	0.20	0.000
Sacramento Deep Water Channel, Light 55	259	7.9	7.12	11.7	240.0	0.27	0.001
Sacramento River at Hood DWR Station	189	7.9	7.30	11.4	107.0	0.22	0.001
Rough and Ready DWR station, Stockton	399	9.5	7.13	10.2	45.1	0.26	0.001
Napa River, near River Park Blvd.	291	9.6	7.45	11.0	195.0	0.27	0.001
Bottle Blank	-	-	-	-	0.3	0.00	-

Table B 6-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 2/01/08 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 1/31/08 - 2/01/08.

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	334	22.8	23.1	7.7	8.7	7.81	8.18	104	72	-
DIEPAMHR + organic matter	337	22.4	22.5	7.4	8.7	7.77	8.11	104	72	-
Old River at mouth of Holland Cut (915)	375	22.7	23.2	7.3	8.6	7.78	8.09	108	74	0.006
San Joaquin River at Potato Slough (815)	243	22.4	23.5	7.3	8.4	7.70	8.07	88	68	0.010
Old River, western arm at railroad bridge (902)	376	22.1	23.0	7.8	8.7	7.81	8.14	108	76	0.007
Sacramento River at tip of Grand Island (711)	133	22.5	23.5	7.7	8.5	7.64	7.97	64	56	0.010
Confluence of Linsey Sl. And Cache Sl.	238	21.4	22.6	7.9	8.9	7.84	8.17	96	82	0.009
Upper Cache Slough at mouth of Ulatis Creek	350	22.4	23.5	7.9	8.6	7.98	8.90	136	106	0.009
Sacramento Deep Water Channel, Light 55	244	22.4	23.5	8.0	8.8	7.88	8.18	96	86	0.013
Sacramento River at Hood DWR Station	162	22.1	22.9	7.8	8.7	7.64	7.98	80	62	0.007
Rough and Ready DWR station, Stockton	378	22.5	24.1	7.8	8.7	7.67	7.99	116	70	0.009
Napa River, near River Park Blvd.	284	22.6	23.8	7.9	8.5	7.66	8.05	100	62	0.011
Bottle Blank	340	22.5	23.7	8.1	8.6	7.80	8.14	120	60	0.000
DIEPAMHR + 25 ppb PBO	332	22.8	23.7	7.5	8.6	7.83	8.16	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	324	22.4	22.8	7.9	8.7	7.65	8.10	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	380	22.4	23.1	7.8	8.8	7.78	8.07	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	235	22.6	23.0	7.6	8.9	7.74	8.12	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	376	22.3	23.1	7.9	8.9	7.79	8.08	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	136	22.5	23.2	7.8	8.8	7.60	8.04	-	-	-
Confluence of Linsey Sl. And Cache Sl. + 25 ppb PBO	234	22.5	23.3	7.9	8.6	7.88	8.14	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	344	22.2	23.3	7.6	8.6	7.95	8.15	-	-	-
Sacramento Deep Water Channel, Light 55 + 25 ppb PBO	238	22.0	23.4	7.9	8.7	7.89	8.19	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	160	21.8	22.4	7.9	8.7	7.70	7.97	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	371	21.8	22.8	7.8	8.9	7.75	7.97	-	-	-
Napa River, near River Park Blvd. + 25 ppb PBO	270	22.4	23.5	7.8	8.6	7.67	8.09	-	-	-

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 B 7-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 2/14/08 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 2/12/08-2/13/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	88	6.3	98	2.5	NS
DIEPAMHR + POM	93	4.8	77	7.3	NS
High EC Control @ 12.46 mS/cm + POM	100	0.0	100	0.0	NS
Suisun at Rush Ranch	100	0.0	100	0.0	NS
Montezuma Slough at Nurse Slough (609)	100	0.0	100	0.0	NS
Napa River at Vallejo Seawall (340)[3]	98	2.5	100	0.0	NS
Grizzly Bay at Dolphin (602)	100	0.0	90	10.0	NS
Suisun Bay off Chipps Island (508)	100	0.0	98	2.5	NS
Carquinez Strait, West of Benicia army dock (405)[3]	93	7.5	98	2.5	NS
Napa River, near River Park Blvd. (NAPA)	98	2.3	98	2.5	NS
Field Duplicate: Suisun Bay off Chipps Island (447)	100	0.0	-	-	NA
Bottle Blank: DIEPAMHR (227)	100	0.0	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.044	0.006	0.048**	0.012	NS
DIEPAMHR + POM	0.059	0.009	0.117	0.015	S*
High EC Control @ 12.46 mS/cm + POM	0.076	0.017	0.085*	0.005	NS
Suisun at Rush Ranch	0.091	0.009	0.132	0.009	S*
Montezuma Slough at Nurse Slough (609)	0.072	0.019	0.113	0.006	NS
Napa River at Vallejo Seawall (340)[3]	0.075	0.010	0.105	0.006	S*
Grizzly Bay at Dolphin (602)	0.086	0.009	0.076*	0.008	NS
Suisun Bay off Chipps Island (508)	0.081	0.004	0.074*	0.013	NS
Carquinez Strait, West of Benicia army dock (405)[3]	0.069	0.005	0.046***	0.003	S*
Napa River, near River Park Blvd. (NAPA)	0.097	0.009	0.086	0.009	NS
Field Duplicate: Suisun Bay off Chipps Island (447)	0.084	0.006	-	-	NA
Bottle Blank: DIEPAMHR (227)	0.087	0.019	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

***: $P < 0.001$

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

3. These high conductivity samples were compared to the High EC Control.

Table B 7-2. Summary of 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 2/12/08-2/13/08.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Suisun at Rush Ranch (SU)	3089	14.1	7.51	9.1	53.4	0.08	0.001
Napa River, Near River Park Blvd. (NAPA)	366	13.0	7.71	10.0	37.6	0.05	0.001
Suisun Bay, off Chipps Island (508)	386	9.4	7.57	10.7	120.3	0.22	0.001
Montezuma Slough at Nurse Slough (609)	3497	10.8	7.27	9.9	103.4	0.24	0.001
Grizzly Bay at Dolphin (602)	364	10.4	7.65	10.2	188.3	0.25	0.002
Napa River at Vallejo Seawall (340)	10250	10.8	7.57	10.4	74.1	0.22	0.001
Field Duplicate: Suisun Bay off Chipps Island (447)	386	9.4	7.57	9.9	111.3	0.22	0.001
Carquinez Strait, West of Benicia army dock (405)	11730	10.3	7.67	10.3	116.7	0.23	0.001

Table B 7-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 2/14/08 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 2/12/08-2/13/08.

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	329	21.7	22.4	7.5	8.7	7.88	8.09	104	58	-
DIEPAMHR + 1% organic matter	329	21.5	22.5	7.4	8.6	7.76	8.12	104	58	-
High EC Control @ 12.46 mS + 1% organic matter	11145	21.8	23.2	7.3	8.4	7.77	8.03	1480	78	-
Suisun at Rush Ranch	2816	21.7	22.7	7.3	8.9	7.74	8.29	400	160	0.002
Montezuma Slough at Nurse Slough (609)	3247	21.7	22.2	7.5	8.8	7.62	7.97	424	80	0.004
Napa River at Vallejo Seawall (340)	9405	22.3	23.5	7.3	8.5	7.84	7.95	1000	90	0.006
Grizzly Bay at Dolphin (602)	336	21.2	22.3	7.5	8.5	7.93	8.10	96	76	0.012
Suisun Bay off Chipps Island (508)	379	22.3	22.4	7.3	8.7	7.91	7.99	96	78	0.009
Carquinez Strait, West of Benicia army dock (405)	10875	21.6	22.3	7.4	8.1	7.82	7.99	1200	90	0.006
Napa River, near River Park Blvd. (NAPA)	343	22.2	22.6	7.5	8.6	7.95	8.10	104	80	0.002
Field Duplicate: Suisun Bay off Chipps Island (447)	415	21.8	22.6	7.4	8.7	7.99	8.06	100	80	0.011
Bottle Blank: DIEPAMHR (227)	1868	22.1	22.5	7.5	8.7	7.83	8.11	116	64	0.000
DIEPAMHR + 25 ppb PBO	335	21.5	22.7	7.7	8.8	7.85	8.05	-	-	-
DIEPAMHR + 1% organic matter + 25 ppb PBO	333	21.6	22.6	7.4	8.6	7.78	8.12	-	-	-
High EC Control @ 12.46 mS + 1% organic matter + 25 ppb PBO	11425	22.0	22.7	7.3	8.3	7.74	8.05	-	-	-
Suisun at Rush Ranch + 25 ppb PBO	2862	22.1	22.2	7.4	8.5	7.97	8.27	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	3225	21.3	23.2	7.5	8.5	7.80	7.96	-	-	-

Napa River at Vallejo Seawall (340) + 25 ppb PBO	9505	21.4	23.0	7.2	8.3	7.81	7.93	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	345	21.2	23.0	7.5	8.5	7.89	8.10	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	378	20.8	23.3	7.3	8.9	7.86	7.99	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	10560	20.3	22.8	7.4	8.5	7.81	7.94	-	-	-
Napa River, near River Park Blvd. (NAPA) + 25 ppb PBO	412	20.2	22.8	7.4	8.8	7.92	8.10	-	-	-

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 B 8-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 2/15/08 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 2/14/08-2/15/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	89	6.1	100	0.0	NS
DIEPAMHR + POM	80	6.9	82*	8.5	NS
Confluence of Lindsey Sl. and Cache Sl. (CL)	100	0.0	98	2.5	NS
Old River, western arm at railroad bridge (902)	95	2.9	97	3.1	NS
Sacramento River, Deep water channel, Light 55	94	3.2	98	2.5	NS
San Joaquin River at Potato Slough (815)	100	0.0	100	0.0	NS
Sacramento River at tip of Grand Island (711)	97	2.8	98	2.5	NS
Upper Cache Slough at mouth of Ulatis Creek (CU)	98	2.5	100	0.0	NS
Old River at mouth of Holland Cut (915)	100	0.0	100	0.0	NS
Sacramento River at Hood DWR Station (Hood)	95	2.9	98	2.5	NS
Rough and Ready DWR Station, Stockton (R&R)	98	2.5	98	2.5	NS
Trip Blank: DIEPAMHR	98	2.5	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.053	0.006	0.056*	0.007	NS
DIEPAMHR + POM	0.055	0.008	0.075	0.005	NS
Confluence of Lindsey Sl. and Cache Sl. (CL)	0.091	0.006	0.091	0.008	NS
Old River, western arm at railroad bridge (902)	0.092	0.009	0.080	0.006	NS
Sacramento River, Deep water channel, Light 55	0.094	0.003	0.070	0.004	S**
San Joaquin River at Potato Slough (815)	0.083	0.009	0.097	0.005	NS
Sacramento River at tip of Grand Island (711)	0.089	0.009	0.090	0.005	NS
Upper Cache Slough at mouth of Ulatis Creek (CU)	0.088	0.005	0.082	0.009	NS
Old River at mouth of Holland Cut (915)	0.090	0.006	0.105	0.008	NS
Sacramento River at Hood DWR Station (Hood)	0.089	0.007	0.095	0.007	NS
Rough and Ready DWR Station, Stockton (R&R)	0.101	0.007	0.085	0.010	NS

Trip Blank: DIEPAMHR	0.057	0.002	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

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

Table B 8-2. Summary of 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 2/14/08-2/15/08

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (^oC)	pH	DO (mg/L)			
Old River, western arm at railroad bridge (902)	314	9.2	7.28	11.8	183.7	0.27	0.001
Old River, at mouth of Holland Cut (915)	333	9.9	7.44	11.4	44.3	0.18	0.001
San Joaquin, just west of Oulton Point (815)	255	9.3	7.24	11.8	44.5	0.17	0.000
Sacramento River, Deep Water Channel, Light 55	362	10.1	7.31	10.9	108.0	0.22	0.001
Sacramento River at tip of Grand Island (711)	341	10.1	7.27	11.4	26.4	0.29	0.001
Confluence of Lindsey Slough and Cache Slough (CL)	292	10.3	7.33	11.2	66.2	0.24	0.001
Upper Cache Slough, Mouth of Ulatis Creek (CU)	570	10.1	7.86	11.3	97.5	0.20	0.002
Sacramento River at Hood DWR Station (HOOD)	393	11.2	7.22	10.4	41.5	0.32	0.001
Rough and Ready DWR Station, Stockton (R&R)	662	11.1	7.26	9.6	19.4	0.18	0.001

Table B 8-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 2/15/08 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 2/14/08-2/15/08.

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	21.8	24.4	7.8	8.4	7.83	8.04	104	58	-
DIEPAMHR + 1% organic matter	345	22.0	23.5	0.9	8.4	7.78	8.02	104	58	-
Confluence of Linsey Sl. and Cache Sl. (CL)	293	22.0	24.2	7.6	8.9	7.58	8.09	104	102	0.009
Old River, western arm at railroad bridge (902)	287	22.0	24.5	7.5	8.8	7.83	7.94	88	60	0.011
Sacramento River, Deep water channel, Light 55	343	22.0	24.0	7.5	8.8	6.15	8.15	116	118	0.014
San Joaquin River at Potato Slough (815)	255	22.0	24.1	7.6	8.6	7.78	7.93	80	100	0.006
Sacramento River at tip of Grand Island (711)	244	22.0	24.4	7.5	8.8	7.91	8.05	84	90	0.016
Upper Cache Slough at mouth of Ulatis Creek (CU)	562	22.0	24.5	7.5	8.8	8.21	8.31	220	164	0.018
Old River at mouth of Holland Cut (915)	306	21.9	23.9	7.3	8.8	7.80	7.97	52	70	0.008
Sacramento River at Hood DWR Station (Hood)	256	22.0	24.6	7.5	8.8	7.83	8.00	92	92	0.011
Rough and Ready DWR Station, Stockton (R&R)	648	22.1	24.5	7.6	8.5	7.88	8.07	244	88	0.010
Trip Blank: DIEPAMHR	345	21.9	24.5	7.7	8.5	7.81	8.06	104	58	0.001
DIEPAMHR	348	22.0	24.0	7.8	8.3	7.83	8.03	-	-	-
DIEPAMHR + 1% organic matter	349	22.0	24.0	7.8	8.4	7.78	8.12	-	-	-
Confluence of Linsey Sl. and Cache Sl. (CL)	293	22.0	24.4	7.6	8.8	7.98	8.12	-	-	-
Old River, western arm at railroad bridge (902)	289	22.0	24.4	7.5	8.9	7.80	7.94	-	-	-
Sacramento River, Deep water channel, Light 55	343	21.9	24.1	7.3	8.7	8.06	8.22	-	-	-
San Joaquin River at Potato Slough (815)	255	22.2	24.2	7.5	8.8	7.83	8.04	-	-	-
Sacramento River at tip of Grand Island (711)	241	22.1	23.7	7.5	8.6	7.91	8.08	-	-	-
Upper Cache Slough at mouth of Ulatis Creek (CU)	556	22.1	24.2	7.5	8.8	8.19	8.33	-	-	-
Old River at mouth of Holland Cut (915)	322	22.1	24.1	7.5	8.5	7.83	7.94	-	-	-
Sacramento River at Hood DWR Station (Hood)	264	22.2	24.4	7.5	8.5	7.95	8.16	-	-	-
Rough and Ready DWR Station, Stockton (R&R)	644	22.2	24.2	7.4	8.8	7.72	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 B 9-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 2/28/08 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 2/26/08-2/27/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	98	2.5	98	2.5	NS
DIEPAMHR + POM	76*	4.4	78	8.5	NS
High EC Control @ 13.81 mS/cm	100	0.0	100	0.0	NS
Suisun @ Rush Ranch	98	2.3	100	0.0	NS
Napa River, near River Park Blvd. (NAPA)	98	2.5	98	2.5	NS
Carquinez Strait, West of Benicia army dock (405)[3]	98	2.5	95	5.0	NS
Napa River at Vallejo Seawall (340)[3]	95	2.9	98	2.5	NS
Suisun Bay off Chipps Island (508)	98	2.5	100	0.0	NS
Montezuma Slough at Nurse Slough (609)	98	2.5	100	0.0	NS
Grizzly Bay at Dolphin (602)	100	0.0	100	0.0	NS
Bottle Blank: DIEPAMHR	92	5.3	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.049	0.007	0.019	0.002	S**
DIEPAMHR + POM	0.051	0.007	0.029	0.005	S*
High EC Control @ 13.81 mS/cm	0.068	0.007	0.059	0.008	NS
Suisun @ Rush Ranch	0.094	0.007	0.084	0.002	NS
Napa River, near River Park Blvd. (NAPA)	0.095	0.009	0.083	0.006	NS
Carquinez Strait, West of Benicia army dock (405)[3]	0.062	0.001	0.038	0.008	NS
Napa River at Vallejo Seawall (340)[3]	0.067	0.007	0.069	0.006	NS
Suisun Bay off Chipps Island (508)	0.112	0.004	0.097	0.005	NS
Montezuma Slough at Nurse Slough (609)	0.081	0.007	0.078	0.010	NS
Grizzly Bay at Dolphin (602)	0.071	0.005	0.065	0.003	NS
Bottle Blank: DIEPAMHR[4]	0.029*	0.008	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

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

3. These high conductivity samples were compared to the High EC control.

4. The bottle blank showed significantly lower weight compared to the POM Control, but not compared to the control without POM.

Table B 9-2. Summary of 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 2/26/08-2/27/08.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Suisun @ Rush Ranch (SU)	3472	13.3	7.61	9.6	60.4	0.11	0.001
Napa River, near River Park Blvd. (NAPA)	260	13.3	7.88	10.6	30.7	0.05	0.001
Suisun Bay, off Chipps Island (508)	473	11.7	7.97	11.2	37.8	0.18	0.003
Montezuma Slough @ Nurse Slough (609)	3626	12.4	7.59	10.7	75.9	0.22	0.002
Grizzly Bay at Dolphin (602)	2090	14.1	7.91	10.8	45.1	0.14	0.002
Napa River at Vallejo Seawall (340)	12970	11.8	7.80	11.0	61.8	0.18	0.002
Carquinez Strait, just west of Benicia army dock (405)	8980	12.0	7.83	10.8	201.7	0.27	0.003

Table B 9-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 2/28/08 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 2/26/08-2/27/08.

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	343	23.4	23.5	7.4	9.4	7.71	8.07	96	56	-
DIEPAMHR + organic matter	349	22.8	23.2	7.1	8.3	7.70	8.03	96	56	-
High EC Control @ 13.81 mS/cm	12855	23.0	23.4	7.5	8.8	7.76	7.84	1760	72	-
Suisun @ Rush Ranch	3295	23.4	23.8	7.3	8.6	7.92	8.31	456	178	0.0
Napa River, near River Park Blvd. (NAPA)	245	23.4	23.7	7.2	8.8	7.78	7.94	76	64	0.0
Carquinez Strait, West of Benicia army dock (405)	9405	23.4	23.9	7.5	8.6	7.84	7.90	1240	94	0.007
Napa River at Vallejo Seawall (340)	12250	23.2	23.6	7.2	8.7	7.81	7.90	1640	94	0.004
Suisun Bay off Chipps Island (508)	500	23.4	23.5	7.4	8.7	7.96	8.15	112	96	0.011
Montezuma Slough at Nurse Slough (609)	3402	23.2	23.5	7.2	8.7	7.72	7.93	408	82	0.005
Grizzly Bay at Dolphin (602)	1950	23.4	23.4	7.4	8.9	7.97	8.05	280	92	0.005
Bottle Blank: DIEPAMHR	348	23.3	23.5	6.9	8.4	7.84	8.07	104	58	0.000
DIEPAMHR + 25 ppb PBO	350	22.9	23.0	7.6	8.3	7.82	8.08	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	339	23.5	24.2	7.0	8.4	7.70	8.01	-	-	-
High EC Control @ 13.81 mS/cm + 25 ppb PBO	12760	23.4	24.0	7.5	8.5	7.77	7.84	-	-	-
Suisun @ Rush Ranch + 25 ppb PBO	3297	23.4	24.3	7.5	8.6	7.77	8.31	-	-	-
Napa River, near River Park Blvd. (NAPA) + 25 ppb PBO	247	23.7	24.5	7.4	8.9	7.75	7.97	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	9515	23.8	24.1	7.4	8.5	7.79	7.92	-	-	-

Napa River at Vallejo Seawall (340) + 25 ppb PBO	12315	23.8	24.2	7.2	8.7	7.73	7.91	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	471	23.7	24.0	7.3	8.9	8.02	8.15	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	3350	23.9	24.0	7.4	8.5	7.73	7.94	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	1958	24.0	24.4	7.5	8.7	7.90	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 B 10-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 2/29/08 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 2/28/08-2/29/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	92	4.8	98	2.5	NS
DIEPAMHR + organic matter	87	4.7	71*	12.8	NS
San Joaquin River at Potato Slough (815)	95	5.0	98	2.5	NS
Confluence of Lindsey Slough and Cache Slough (CL)	95	2.8	100	0.0	NS
Sacramento River, Deep Water Channel, Light 55	98	2.5	95	2.9	NS
Sacramento River at tip of Grand Island (711)	95	2.9	100	0.0	NS
Upper Cache Slough at mouth of Ulatis Creek (CU)	98	2.5	66*	9.3	S* (67%)
Old River, western arm at railroad bridge (902)	98	2.5	100	0.0	NS
Old River at mouth of Holland Cut (915)	100	0.0	100	0.0	NS
Rough and Ready DWR Station, Stockton (R&R)	100	0.0	100	0.0	NS
Sacramento River at Hood DWR Station	98	2.5	97	2.8	NS
Field Duplicate: Sacramento River at tip of Grand Island (447)	98	2.5	-	-	NA
Trip Blank: DIEPAMHR	93	2.5	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.022*	0.007	0.049	0.003	S* (223%)
DIEPAMHR + organic matter	0.050	0.010	0.057	0.010	NS
San Joaquin River at Potato Slough (815)	0.092	0.009	0.103	0.003	NS
Confluence of Lindsey Slough and Cache Slough (CL)	0.103	0.004	0.106	0.007	NS
Sacramento River, Deep Water Channel, Light 55	0.078	0.010	0.098	0.003	NS
Sacramento River at tip of Grand Island (711)	0.055	0.007	0.060	0.005	NS
Upper Cache Slough at mouth of Ulatis Creek (CU)	0.080	0.008	0.051	0.013	NS
Old River, western arm at railroad bridge (902)	0.107	0.009	0.091	0.010	NS
Old River at mouth of Holland Cut (915)	0.103	0.003	0.105	0.006	NS
Rough and Ready DWR Station, Stockton (R&R)	0.101	0.008	0.129	0.007	S* (128%)

Sacramento River at Hood DWR Station	0.069	0.003	0.090	0.014	NS
Field Duplicate: Sacramento River at tip of Grand Island (447)	0.076	0.005	-	-	NA
Trip Blank: DIEPAMHR	0.049	0.010	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard statistical protocols.

*: $P < 0.05$

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

Table B 10-2. Summary of 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 2/28/08-2/29/08.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (^{o}C)	pH	DO (mg/L)			
Old River, Western arm at railroad bridge (902)	314	12.5	7.85	10.8	19.0	0.11	0.002
Old River at mouth of Holland Cut (915)	340	12.5	7.81	10.5	14.6	0.14	0.002
San Joaquin, just west of Oulton Point (815)	273	11.8	7.76	10.8	14.6	0.24	0.003
Sacramento River, Deep Water Channel, Light 55	303	11.1	7.79	10.9	76.7	0.22	0.003
Sacramento River at tip of Grand Island (711)	148	10.8	7.82	11.0	243.0	0.38	0.005
Confluence of Lindsey Slough at Cache Slough (CL)	323	11.0	7.91	11.1	76.6	0.21	0.003
Upper Cache Slough, Mouth of Ulatis Creek (CU)	575	12.6	8.00	9.7	59.8	0.17	0.003
Field Duplicate: Sacramento River at tip of Grand Island (447)	148	10.8	7.82	11.0	241.0	0.00	0.000
Sacramento River at Hood DWR Station (HOOD)	165	12.3	7.91	10.7	99.8	0.37	0.006
Rough and Ready DWR Station, Stockton (R&R)	829	13.7	7.82	9.9	10.7	0.19	0.003

Table B 10-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 2/29/08 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 2/28/28-2/29/08.

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	24.4	7.2	8.5	7.69	8.03	96	56	-
DIEPAMHR + organic matter	350	23.0	24.3	7.4	8.3	7.70	7.94	96	56	-
San Joaquin River at Potato Slough (815)	263	23.1	24.8	7.4	8.6	7.84	8.04	92	86	0.009
Confluence of Lindsey Slough and Cache Slough (CL)	267	23.0	24.9	7.4	8.9	7.89	8.04	88	88	0.009
Sacramento River, Deep Water Channel, Light 55	324	23.1	24.8	7.5	8.9	7.93	8.11	104	102	0.009
Sacramento River at tip of Grand Island (711)	145	23.1	24.4	7.4	8.9	7.70	7.91	56	56	0.010
Upper Cache Slough at mouth of Ulatis Creek (CU)	533	23.1	24.7	7.5	8.8	8.04	8.27	172	148	0.009
Old River, western arm at railroad bridge (902)	308	23.0	24.0	7.4	8.5	7.83	8.03	98	84	0.004
Old River at mouth of Holland Cut (915)	334	23.1	24.6	7.5	8.6	7.76	8.00	102	80	0.005
Rough and Ready DWR Station, Stockton (R&R)	810	23.2	24.7	7.3	8.8	7.87	8.08	184	114	0.003
Sacramento River at Hood DWR Station	159	23.2	24.9	7.4	8.6	7.65	7.79	68	62	0.006
Field Duplicate: Sacramento River at tip of Grand Island (447)	159	23.2	24.3	7.5	8.6	7.67	7.86	60	58	0.013
Trip Blank: DIEPAMHR	254	23.1	24.8	7.5	8.4	7.66	8.01	104	58	0.000
DIEPAMHR + 25 ppb PBO	346	23.2	24.1	7.6	8.6	7.73	7.98	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	345	23.1	24.4	7.1	8.5	7.55	7.95	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	310	23.2	24.7	7.2	8.9	7.82	8.05	-	-	-
Confluence of Lindsey Slough and Cache Slough (CL) + 25 ppb PBO	265	23.4	23.7	7.5	8.9	7.87	8.06	-	-	-
Sacramento River, Deep Water Channel, Light 55 + 25 ppb PBO	289	23.2	24.1	7.5	8.9	7.95	8.12	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	232	23.2	23.4	7.5	8.9	7.69	7.87	-	-	-
Upper Cache Slough at mouth of Ulatis Creek (CU) + 25 ppb PBO	343	23.1	24.5	7.4	8.6	8.01	8.27	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	423	23.3	24.6	7.4	8.6	7.83	8.06	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	322	23.6	24.9	7.6	8.8	7.82	8.04	-	-	-
Rough and Ready DWR Station, Stockton (R&R) + 25 ppb PBO	560	23.2	24.6	7.3	8.9	7.77	8.12	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	469	23.4	24.8	7.5	8.5	7.80	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 B 11-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 3/13/08 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 3/11/08-3/12/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	100	0.0	98	2.3	NS
Suisun @ Rush Ranch	100	0.0	100	0.0	NS
Napa River, near River Park Blvd. (NAPA)	95	5.0	100	0.0	NS
Rough and Ready DWR Station, Stockton (R&R)	98	2.5	98	2.5	NS
Sacramento River at Hood DWR Station (HOOD)	95	5.0	98	2.5	NS
Trip Blank: DIEPAMHR	98	2.5	-	-	NA
Field Duplicate: Sacramento River at Hood DWR Station (447)	88**	2.5	-	-	NA
Bottle Blank: DIEPAMHR	98	2.5	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.052	0.004	0.047	0.004	NS
Suisun @ Rush Ranch	0.088	0.003	0.088	0.009	NS
Napa River, near River Park Blvd. (NAPA)	0.082	0.004	0.080	0.004	NS
Rough and Ready DWR Station, Stockton (R&R)	0.093	0.008	0.073	0.010	NS
Sacramento River at Hood DWR Station (HOOD)	0.060	0.006	0.067	0.007	NS
Trip Blank: DIEPAMHR	0.035**	0.002	-	-	NA
Field Duplicate: Sacramento River at Hood DWR Station (447)	0.071	0.004	-	-	NA
Bottle Blank: DIEPAMHR	0.054	0.004	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard statistical protocols.

**: $P < 0.01$

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

Table B 11-2. Summary of 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 3/11/08-3/12/08.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (^{o}C)	pH	DO (mg/L)			
Suisun at Rush Ranch (SU)	3928	16.3	7.57	8.6	50.0	0.11	0.001
Napa River, near River Park Blvd. (NAPA)	553	14.9	7.89	10.6	7.3	0.03	0.001
Sacramento River at Hood DWR Station (HOOD)	357	14.5	7.60	9.5	13.2	0.40	0.004
Rough and Ready DWR Station, Stockton (R&R)	907	15.9	7.78	9.3	13.8	0.08	0.001
Trip Blank: DIEPAMHR (337)	357	21.2	8.09	9.1	0.2	0.01	0.000
Field Duplicate: Sacramento River at Hood DWR Station (447)	357	14.5	7.60	9.5	14.6	0.39	0.004

Table B 11-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 3/13/08 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 3/11/08-3/12/08.

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 (^{o}C)	Max Temp (^{o}C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	347	23.1	24.4	7.6	8.6	7.72	8.11	108	60	-
Suisun @ Rush Ranch	3572	22.2	25.0	7.6	8.3	7.94	8.47	512	186	0.004
Napa River, near River Park Blvd. (NAPA)	468	22.0	24.2	7.7	8.4	7.98	8.31	140	106	0.002
Rough and Ready DWR Station, Stockton (R&R)	833	22.2	24.3	7.4	8.5	7.96	8.24	192	122	0.003
Sacramento River at Hood DWR Station (HOOD)	215	22.4	24.5	7.7	8.6	7.82	8.07	84	82	0.023
Trip Blank: DIEPAMHR	332	22.6	24.4	7.8	8.4	7.70	8.08	112	58	0.001
Field Duplicate: Sacramento River at Hood DWR Station (447)	214	22.4	24.7	7.5	8.5	7.76	8.02	80	84	0.011
Bottle Blank: DIEPAMHR	332	22.3	24.4	7.8	8.8	7.73	8.10	112	60	0.000
DIEPAMHR + 25 ppb PBO	339	22.4	24.6	7.8	8.5	7.76	8.23	-	-	-
DIEPAMHR + Glassware contamination test + 25 ppb PBO	334	21.7	24.4	7.8	8.6	7.27	8.10	-	-	-
Suisun @ Rush Ranch + 25 ppb PBO	3346	22.6	24.6	7.5	8.9	7.86	8.50	-	-	-
Napa River, near River Park Blvd. (NAPA) + 25 ppb PBO	520	22.6	24.4	7.7	9.0	7.97	8.41	-	-	-
Rough and Ready DWR Station, Stockton (R&R) + 25 ppb PBO	831	22.4	24.6	6.7	8.6	7.98	8.24	-	-	-
Sacramento River at Hood DWR Station (HOOD) + 25 ppb PBO	207	22.2	24.5	6.9	8.9	7.52	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 B 12-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 3/15/08 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 3/13/08-3/14/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	94	3.2	100	0.0	NS
High EC Control @ 17.09 mS/cm	98	2.5	98	2.5	NS
High EC Control @ 23.45 mS/cm	85	8.7	55*	14.4	NS
Confluence of Lindsey Slough and Cache Slough (CL)	98	2.3	100	0.0	NS
Old River, western arm at railroad bridge (902)	100	0.0	100	0.0	NS
Upper Cache Slough at mouth of Ulatis Creek (CU)	97	2.8	100	0.0	NS
Old River at mouth of Holland Cut (915)	100	0.0	97	2.8	NS
San Joaquin River at Potato Slough (815)	97	2.8	98	2.5	NS
Sacramento River at tip of Grand Island (711)	89	0.3	92	2.6	NS
Sacramento River, Deep Water Channel, Light 55	100	0.0	95	2.9	NS
Suisun Bay off Chipps Island (508)[4]	100	0.0	97	3.1	NS
Carquinez Strait, West of Benicia army dock (405)[3]	98	2.5	89	4.1	NS
Grizzly Bay at Dolphin (602)	100	0.0	98	2.5	NS
Montezuma Slough at Nurse Slough (609)	97	2.8	100	0.0	NS
Napa River at Vallejo Seawall (340)[4]	71	6.4	82	11.0	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.037	0.003	0.041	0.008	NS
High EC Control @ 17.09 mS/cm	0.040	0.003	0.022*	0.004	S** (55%)
High EC Control @ 23.45 mS/cm	0.029	0.008	0.008**	0.003	S* (28%)
Confluence of Lindsey Slough and Cache Slough (CL)	0.069	0.009	0.064	0.005	NS
Old River, western arm at railroad bridge (902)	0.072	0.004	0.062	0.005	NS
Upper Cache Slough at mouth of Ulatis Creek (CU)	0.078	0.005	0.048	0.003	S** (62%)
Old River at mouth of Holland Cut (915)	0.070	0.007	0.070	0.003	NS
San Joaquin River at Potato Slough (815)	0.060	0.007	0.067	0.005	NS
Sacramento River at tip of Grand Island (711)	0.051	0.006	0.061	0.003	NS
Sacramento River, Deep Water Channel, Light 55	0.075	0.007	0.053	0.004	S* (71%)
Suisun Bay off Chipps Island (508)[4]	0.072	0.005	0.055	0.011	NS
Carquinez Strait, West of Benicia army dock (405)[3]	0.051	0.001	0.051	0.005	NS
Grizzly Bay at Dolphin (602)	0.049	0.006	0.067	0.005	NS
Montezuma Slough at Nurse Slough (609)	0.067	0.009	0.064	0.007	NS

Napa River at Vallejo Seawall (340)[4]	0.034	0.004	0.042	0.003	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

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

3. This high conductivity sample was compared to the High EC Control @ 17.09 mS

4. These high conductivity samples were compared to the High EC Control @ 23.45 mS.

Table B 12-2. Summary of 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 3/13/08-3/14/08

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Confluence of Lindsey Slough and Cache Slough (CL)	349	14.2	8.13	10.0	48.9	0.23	0.007
Old River, western arm at railroad bridge (902)	382	14.9	8.24	9.8	19.2	0.08	0.003
Upper Cache Slough at mouth of Ulatis Creek (CU)	721	14.6	8.43	10.0	32.7	0.06	0.004
Old River at mouth of Holland Cut (915)	361	14.9	8.16	10.0	16.6	0.08	0.003
San Joaquin River at Potato Slough (815)	280	14.1	8.25	10.1	14.7	0.15	0.006
Sacramento River at tip of Grand Island (711)	237	14.2	8.16	10.1	9.3	0.39	0.013
Sacramento River, Deep Water Channel, Light 55	366	14.1	8.09	10.0	30.0	0.22	0.006
Suisun Bay, off Chipps Island (508)	2274	13.2	8.15	10.5	55.0	0.12	0.003
Carquinez Strait, west of Benicia army dock (405)	16190	12.9	8.05	10.9	16.3	0.11	0.002
Grizzly Bay at Dolphin (602)	8230	13.0	8.11	10.5	32.7	0.13	0.003
Montezuma Slough at Nurse Slough (609)	3651	13.8	7.92	9.6	7.7	0.21	0.003
Napa River at Vallejo Seawall (340)	20020	13.1	8.02	10.6	12.6	0.11	0.002

Table B 12-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 3/15/08 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 3/13/08-3/14/08.

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	22.9	23.4	6.5	8.5	7.54	8.90	108	60	-
High EC Control @ 17.09 mS	15430	22.2	23.7	6.7	8.5	7.58	7.91	2000	80	-
High EC Control @ 23.45 mS	20880	22.7	23.8	6.7	8.4	7.62	7.95	2680	88	-
Confluence of Lindsey Slough and Cache Slough (CL)	375	22.6	23.7	6.2	8.8	7.73	8.24	124	124	0.016
Old River, western arm at railroad bridge (902)	301	23.5	24.1	6.5	8.6	7.61	8.15	100	84	0.005
Upper Cache Slough at mouth of Ulatis Creek (CU)	664	23.5	24.0	6.9	8.6	8.10	8.53	228	200	0.004
Old River at mouth of Holland Cut (915)	335	22.3	23.9	6.5	8.7	7.60	8.15	108	84	0.003
San Joaquin River at Potato Slough (815)	268	23.3	24.1	6.3	8.8	7.56	8.10	96	82	0.008
Sacramento River at tip of Grand Island (711)	248	23.0	24.1	6.4	8.5	7.47	8.16	88	88	0.017
Sacramento River, Deep Water Channel, Light 55	343	23.3	24.3	6.5	8.6	7.84	8.28	132	122	0.017
Suisun Bay off Chipps Island (508)	2082	22.5	23.9	7.4	8.7	7.83	8.14	280	90	0.005
Carquinez Strait, West of Benicia army dock (405)	15390	23.3	24.1	6.8	8.6	7.67	8.04	1840	96	0.002
Grizzly Bay at Dolphin (602)	3959	22.5	24.3	6.8	8.5	7.54	8.08	980	94	0.003
Montezuma Slough at Nurse Slough (609)	1861	22.6	24.5	7.1	8.6	7.66	8.08	432	92	0.008
Napa River at Vallejo Seawall (340)	20430	23.6	24.5	6.6	8.5	7.51	8.05	2560	100	0.002
DIEPAMHR + 25 ppb PBO	336	23.1	23.4	6.9	8.5	7.71	8.04	-	-	-
High EC Control @ 17.09 mS + 25 ppb PBO	15130	22.3	23.9	6.9	8.5	7.61	7.99	-	-	-
High EC Control @ 23.45 mS + 25 ppb PBO	20715	22.9	24.1	7.1	8.3	7.67	8.02	-	-	-
Confluence of Lindsey Slough and Cache Slough (CL) + 25 ppb PBO	343	22.0	23.7	6.6	8.5	7.78	8.26	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	291	21.5	24.0	6.5	8.6	7.61	8.12	-	-	-
Upper Cache Slough at mouth of Ulatis Creek (CU) + 25 ppb PBO	655	22.0	24.1	6.8	8.7	7.77	8.56	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	320	21.1	23.8	6.9	8.6	7.69	8.09	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	269	20.9	24.2	6.4	8.8	7.70	8.15	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	239	21.2	24.2	6.6	9.0	7.72	8.12	-	-	-
Sacramento River, Deep Water Channel, Light 55 + 25 ppb PBO	342	21.2	24.5	6.5	8.8	7.87	8.30	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	2049	21.7	24.7	7.6	8.8	7.60	8.24	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	15030	21.6	24.4	7.3	8.3	7.75	8.09	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	7835	21.1	24.0	7.3	8.3	7.77	8.12	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	3327	21.8	24.3	7.3	8.7	7.80	8.21	-	-	-
Napa River at Vallejo Seawall (340) + 25 ppb PBO	19785	21.2	23.7	6.9	8.4	7.70	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 B 13-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 3/27/08 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 3/25/08 - 3/26/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	100	0.0	97	2.8	NS
Sacramento River at Hood DWR Station	95	5.0	100	0.0	NS
Rough and Ready DWR station, Stockton	100	0.0	100	0.0	NS
Sacramento R. Deep Water Channel, Light 55	100	0.0	100	0.0	NS
Confluence of Linsey Sl. And Cache Sl. (CL)	98	2.5	100	0.0	NS
Upper Cache Slough at mouth of Ulatis Creek (CU)	98	2.3	98	2.5	NS
Suisun Bay at Public Suisun Dock	100	0.0	100	0.0	NS
Napa River at River Park Blvd.	100	0.0	100	0.0	NS
Old River, western arm at railroad bridge (902)	100	0.0	100	0.0	NS
Sacramento River at tip of Grand Island (711)	100	0.0	98	2.5	NS
San Joaquin River at Potato Slough (815)	98	2.3	98	2.5	NS
Old River at mouth of Holland Cut (915)	100	0.0	100	0.0	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.053	0.002	0.046	0.005	NS
Sacramento River at Hood DWR Station	0.084	0.011	0.091	0.006	NS
Rough and Ready DWR station, Stockton	0.127	0.010	0.099	0.018	NS
Sacramento R. Deep Water Channel, Light 55	0.091	0.006	0.077	0.007	NS
Confluence of Linsey Sl. And Cache Sl.	0.092	0.007	0.076	0.008	NS
Upper Cache Slough at mouth of Ulatis Creek	0.108	0.008	0.077	0.007	S* (71%)
Suisun Bay at Public Suisun Dock	0.084	0.009	0.085	0.010	NS
Napa River at River Park Blvd.	0.105	0.017	0.086	0.013	NS
Old River, western arm at railroad bridge (902)	0.101	0.006	0.093	0.012	NS
Sacramento River at tip of Grand Island (711)	0.064	0.009	0.085	0.016	NS
San Joaquin River at Potato Slough (815)	0.101	0.006	0.093	0.006	NS
Old River at mouth of Holland Cut (915)	0.096	0.009	0.109	0.013	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard statistical protocols.

*: $P < 0.05$

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

Table B 13-2. Summary of 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 03/25/08 - 03/26/08.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (^{o}C)	pH	DO (mg/L)			
Sacramento River at Hood DWR Station	207	17.1	7.91	8.9	9.2	0.43	0.010
Rough and Ready DWR Station, Stockton	1045	16.8	8.10	9.2	13.1	0.07	0.003
Sacramento River, Deep Water Channel, Light 55	364	14.8	8.18	10.2	51.9	0.18	0.007
Confluence of Lindsey Slough and Cache Slough (CL)	459	15.1	8.25	10.1	36.5	0.21	0.009
Upper Cache Slough at mouth of Ulatis Creek (CU)	826	14.9	8.43	10.2	21.8	0.00	0.000
Suisun Bay at Public Suisun Dock (SU)	3759	12.8	7.97	8.6	74.2	0.07	0.001
Napa River at River Park Blvd.	714	15.6	8.70	9.8	16.5	0.00	0.000
Old River, western arm at railroad bridge (902)	353	16.0	8.20	10.1	12.3	0.00	0.000
Sacramento River at tip of Grand Island (711)	234	15.4	8.10	10.0	7.4	0.49	0.016
San Joaquin River at Potato Slough (815)	324	15.3	8.31	10.3	8.0	0.06	0.003
Old River at mouth of Holland Cut (915)	398	16.0	8.18	10.0	11.4	0.00	0.000

Table B 13-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 3/27/08 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 3/25/08-3/26/08.

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 (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	322	21.5	22.2	7.9	8.5	7.80	8.17	108	62	-
Sacramento River at Hood DWR Station	185	21.4	22.3	7.7	8.7	7.84	8.06	84	78	0.019
Rough and Ready DWR station, Stockton	809	21.5	23.2	7.9	8.7	8.05	8.21	200	126	0.004
Sacramento R. Deep Water Channel, Light 55	330	21.4	22.0	7.7	8.5	8.06	8.25	132	124	0.011
Confluence of Linsey Sl. And Cache Sl. (CL)	328	21.4	21.9	7.8	8.6	8.05	8.20	128	126	0.013
Upper Cache Slough at mouth of Ulatis Creek (CU)	706	21.1	22.7	7.8	8.7	8.34	8.44	248	222	0.000
Suisun Bay at Public Suisun Dock	3464	21.2	23.0	7.7	8.8	7.96	8.32	528	184	0.002
Napa River at River Park Blvd.	639	21.3	22.8	7.8	8.7	8.02	8.25	172	124	0.000
Old River, western arm at railroad bridge (902)	281	21.3	22.6	7.8	8.9	7.91	8.18	104	84	0.000
Sacramento River at tip of Grand Island (711)	180	21.3	23.2	7.8	8.5	7.83	7.99	92	82	0.022
San Joaquin River at Potato Slough (815)	259	21.3	22.9	7.7	8.8	7.87	8.11	66	84	0.003
Old River at mouth of Holland Cut (915)	309	21.2	23.2	7.8	8.7	7.87	8.04	116	86	0.000
DIEPAMHR + 25 ppb PBO	311	21.1	22.7	7.8	8.5	7.77	8.15	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	179	21.1	21.9	7.6	8.8	7.78	8.09	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	806	21.1	22.3	7.8	8.7	8.06	8.18	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	572	21.0	23.0	7.8	8.9	8.08	8.22	-	-	-
Confluence of Linsey Sl. And Cache Sl. (CL) + 25 ppb PBO	325	21.1	21.8	7.7	8.4	8.05	8.26	-	-	-
Upper Cache Slough at mouth of Ulatis Creek (CU) + 25 ppb PBO	703	21.4	22.0	7.7	8.8	8.32	8.44	-	-	-
Suisun Bay at Public Suisun Dock + 25 ppb PBO	3349	20.9	21.9	7.7	8.2	7.99	8.27	-	-	-
Napa River at River Park Blvd. + 25 ppb PBO	658	21.4	22.5	7.8	8.6	8.05	8.26	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	287	20.9	22.3	7.9	8.6	7.90	8.19	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	184	22.3	22.7	7.6	8.5	7.83	8.07	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	254	20.9	22.3	7.8	8.5	7.89	8.10	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	311	21.0	22.6	7.8	8.7	7.87	8.09	-	-	-

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 B 14-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 3/28/08 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 3/27/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	100	0.0	98	2.5	NS
High EC Control @ 14.20 mS/cm	100	0.0	100	0.0	NS
High EC Control @ 18.76 mS/cm	100	0.0	93	4.8	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	93	4.8	NS
Napa River at Vallejo Seawall (340)[3]	94	3.2	94	5.6	NS
Carquinez Strait, West of Benicia army dock (405)[4]	87**	2.2	97	2.8	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.046	0.008	0.055	0.004	NS
High EC Control @ 14.20 mS/cm	0.037	0.001	0.042	0.012	NS
High EC Control @ 18.76 mS/cm	0.040	0.002	0.032**	0.002	S* (80%)
Suisun Bay off Chipps Island (508)	0.100	0.005	0.077	0.007	S* (77%)
Grizzly Bay at Dolphin (602)	0.070	0.010	0.078	0.005	NS
Montezuma Slough at Nurse Slough (609)	0.074	0.002	0.078	0.005	NS
Napa River at Vallejo Seawall (340)[3]	0.047	0.001	0.066	0.002	S* (140%)
Carquinez Strait, West of Benicia army dock (405)[4]	0.073	0.006	0.067	0.012	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

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

3. This high EC sample was compared to the High EC Control @ 14.20 mS.

4. This high EC sample was compared to the High EC Control @ 18.76 mS.

Table B 14-2. Summary of 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 03/27/08.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (^{o}C)	pH	DO (mg/L)			
Suisun Bay off Chipps Island (508)	1731	13.4	8.24	10.8	43.1	0.14	0.005
Grizzly Bay at Dolphin (602)	3921	13.6	8.28	11.1	40.8	0.11	0.004
Montezuma Slough at Nurse Slough (609)	3838	14.5	8.04	10.4	77.1	0.09	0.002
Napa River at Vallejo Seawall (340)	18580	12.9	8.03	10.6	28.8	0.09	0.002
Carquinez Strait, West of Benicia army dock (405)	13010	13.4	8.04	10.7	157.7	0.22	0.004

Table B 14-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 3/28/08 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 3/27/08.

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 (^{o}C)	Max Temp (^{o}C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	329	22.1	23.0	7.4	8.4	7.81	8.06	108	62	-
High EC Control @ 14.20 mS/cm	13085	22.2	23.8	7.8	8.4	7.77	7.93	1600	76	-
High EC Control @ 18.76 mS/cm	16900	22.4	24.2	7.4	8.1	7.72	7.92	2200	82	-
Suisun Bay off Chipps Island (508)	1842	22.4	22.7	7.8	8.5	7.99	8.14	248	94	0.006
Grizzly Bay at Dolphin (602)	4469	22.4	24.2	7.7	8.7	7.94	8.04	640	96	0.005
Montezuma Slough at Nurse Slough (609)	3535	22.3	24.3	7.8	8.7	7.83	8.04	464	92	0.003
Napa River at Vallejo Seawall (340)	16615	22.3	23.6	7.3	8.3	7.73	7.95	1880	108	0.003
Carquinez Strait, West of Benicia army dock (405)	12420	22.3	23.8	7.4	8.4	7.83	7.98	1680	100	0.008
DIEPAMHR + 25 ppb PBO	363	22.3	23.1	7.6	8.4	7.83	8.08	-	-	-
High EC Control @ 14.20 mS/cm + 25 ppb PBO	12980	22.3	23.1	7.5	8.5	7.79	7.92	-	-	-
High EC Control @ 18.76 mS/cm + 25 ppb PBO	9377	22.3	23.5	7.5	8.1	7.71	7.91	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	9172	22.3	23.3	7.8	8.6	7.94	8.15	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	4487	22.5	23.4	7.5	8.7	7.89	8.00	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	3471	22.4	23.3	7.7	8.4	7.84	8.03	-	-	-
Napa River at Vallejo Seawall (340) + 25 ppb PBO	16595	22.3	23.7	7.2	8.3	7.76	7.92	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	12630	22.4	23.5	7.5	8.5	7.81	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 B 15-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 4/10/08 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 4/08/08 - 4/09/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	100	0.0	93	2.5	NS
Sacramento River at Hood DWR Station	93	2.5	95	5.0	NS
Rough and Ready DWR Station, Stockton	100	0.0	98	2.5	NS
Sacramento River, Deep Water Channel, Light 55	95	2.9	100	0.0	NS
Confluence of Lindsey Slough and Cache Slough (CL)	100	0.0	98	2.5	NS
Upper Cache Slough at mouth of Ulatis Creek (CU)	100	0.0	98	2.5	NS
Suisun Slough at Rush Ranch	95	2.9	100	0.0	NS
Napa River at River Park Blvd.	100	0.0	100	0.0	NS
Old River, western arm at railroad bridge (902)	100	0.0	100	0.0	NS
Sacramento River at tip of Grand Island (711)	100	0.0	88	9.5	NS
San Joaquin River at Potato Slough (815)	98	2.5	95	2.9	NS
Old River at mouth of Holland Cut (915)	100	0.0	98	2.5	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.035	0.003	0.031	0.005	NS
Sacramento River at Hood DWR Station	0.044	0.007	0.042	0.001	NS
Rough and Ready DWR Station, Stockton	0.074	0.007	0.064	0.006	NS
Sacramento River, Deep Water Channel, Light 55	0.039	0.008	0.052	0.005	NS
Confluence of Lindsey Slough and Cache Slough (CL)	0.057	0.008	0.054	0.008	NS
Upper Cache Slough at mouth of Ulatis Creek (CU)	0.056	0.004	0.034	0.004	S* (61%)
Suisun Slough at Rush Ranch	0.058	0.011	0.058	0.005	NS

Napa River at River Park Blvd.	0.058	0.009	0.061	0.009	NS
Old River, western arm at railroad bridge (902)	0.069	0.012	0.062	0.007	NS
Sacramento River at tip of Grand Island (711)	0.044	0.007	0.036	0.007	NS
San Joaquin River at Potato Slough (815)	0.045	0.004	0.042	0.006	NS
Old River at mouth of Holland Cut (915)	0.070	0.009	0.068	0.008	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

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

Table B 15-2. Summary of 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 4/8/08-4/9/08.

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 Hood DWR Station	221	16.6	8.05	9.3	8.1	0.59	0.019
Rough and Ready DWR Station, Stockton	716	16.1	7.97	9.8	11.1	0.07	0.002
Sacramento River, Deep Water Channel, Light 55	396	14.4	8.24	10.6	28.3	0.15	0.006
Confluence of Lindsey Slough and Cache Slough (CL)	359	14.4	8.23	10.5	29.4	0.21	0.008
Upper Cache Slough at mouth of Ulatis Creek (CU)	498	14.3	8.31	10.7	47.3	0.03	0.001
Suisun Slough at Rush Ranch	4603	14.5	7.15	8.6	84.6	0.04	0.000
Napa River at River Park Blvd.	5110	15.3	8.52	12.4	21.1	0.00	0.000
Old River, western arm at railroad bridge (902)	340	15.8	8.30	10.5	14.2	0.00	0.000
Sacramento River at tip of Grand Island (711)	219	15.7	8.06	10.1	8.2	0.50	0.015
San Joaquin River at Potato Slough (815)	317	15.3	8.29	9.5	7.2	0.01	0.000
Old River at mouth of Holland Cut (915)	387	16.1	8.20	10.4	10.3	0.00	0.000

Table B 15-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 4/10/08 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 4/8/08-4/9/08.

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	19.2	24.1	7.6	8.3	7.59	8.07	92	64	0.000
Sacramento River at Hood DWR Station	187	19.7	24.3	7.5	8.3	7.75	7.94	84	98	0.024
Rough and Ready DWR Station, Stockton	665	20.6	24.3	7.7	8.4	7.90	8.01	156	96	0.003
Sacramento River, Deep Water Channel, Light 55	363	20.4	24.1	7.6	8.4	8.03	8.17	124	119	0.009
Confluence of Lindsey Slough and Cache Slough (CL)	330	22.2	24.2	7.3	8.5	8.01	8.13	120	117	0.013
Upper Cache Slough at mouth of Ulatis Creek (CU)	446	22.4	24.1	7.8	8.5	8.13	8.27	166	158	0.002
Suisun Slough at Rush Ranch	3882	22.4	24.0	7.4	8.3	7.84	8.22	564	168	0.001
Napa River at River Park Blvd.	4435	22.2	24.2	7.5	8.6	7.85	8.27	592	128	0.000
Old River, western arm at railroad bridge (902)	296	22.4	24.1	7.8	8.8	7.97	8.07	100	86	0.000
Sacramento River at tip of Grand Island (711)	187	22.3	24.2	6.8	8.9	7.84	8.10	76	78	0.030
San Joaquin River at Potato Slough (815)	279	22.5	24.3	6.6	8.8	7.86	8.09	92	80	0.001
Old River at mouth of Holland Cut (915)	349	22.3	24.2	6.5	8.8	7.95	8.05	112	88	0.000
DIEPAMHR + 25 ppb PBO	316	22.4	23.7	7.7	8.4	7.74	8.07	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	190	22.4	23.7	7.2	8.6	7.78	8.00	-	-	-
Rough and Ready DWR Station, Stockton + 25 ppb PBO	649	22.4	23.8	7.6	8.4	7.88	8.11	-	-	-
Sacramento River, Deep Water Channel, Light 55 + 25 ppb PBO	350	22.5	23.7	7.2	8.6	8.02	8.15	-	-	-
Confluence of Lindsey Slough and Cache Slough (CL) + 25 ppb PBO	326	22.4	23.7	7.5	8.3	8.01	8.14	-	-	-
Upper Cache Slough at mouth of Ulatis Creek (CU) + 25 ppb PBO	441	22.4	23.9	7.8	8.4	8.13	8.26	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	3819	22.3	23.8	7.3	8.3	7.85	8.20	-	-	-
Napa River at River Park Blvd. + 25 ppb PBO	4350	22.5	23.7	7.3	8.8	7.87	8.30	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	297	22.7	24.0	6.6	8.4	7.94	8.21	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	195	22.5	24.0	6.7	8.6	7.81	8.11	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	282	23.0	24.1	6.7	8.3	7.86	8.13	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	350	22.9	23.6	6.2	8.6	7.92	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 B 16-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 04/11/08 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 04/10/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	97	2.8	100	0.0	NS
High EC Control @ 17.54 mS	83	7.5	50*	11.9	NS
High EC Control @ 23.43 mS	42*	8.2	21*	8.0	NS
Suisun Bay off Chipps Island (508)	98	2.5	100	0.0	NS
Grizzly Bay at Dolphin (602)	98	2.3	95	5.0	NS
Montezuma Slough at Nurse Slough (609)	100	0.0	95	5.0	NS
Napa River at Vallejo Seawall (340)[3]	89	6.1	71	6.5	NS
Carquinez Strait, West of Benicia army dock (405)[4]	94	5.6	93	4.1	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.042	0.006	0.054	0.008	NS
High EC Control @ 17.54 mS	0.024	0.014	0.043	0.009	NS
High EC Control @ 23.43 mS	0.039	0.004	0.066	0.024	NS
Suisun Bay off Chipps Island (508)	0.054	0.007	0.057	0.006	NS
Grizzly Bay at Dolphin (602)	0.042	0.005	0.037	0.009	NS
Montezuma Slough at Nurse Slough (609)	0.067	0.010	0.065	0.005	NS
Napa River at Vallejo Seawall (340)[3]	0.050	0.007	0.021**	0.003	NS
Carquinez Strait, West of Benicia army dock (405)[4]	0.050	0.007	0.044	0.005	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

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

3. This high EC sample was compared to the High EC Control @ 23.43 mS.

4. This high EC sample was compared to the High EC Control @ 17.54 mS.

Table B 16-2. Summary of 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 4/10/08.

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)	5970	14.2	8.07	10.9	22.1	0.05	0.001
Grizzly Bay at Dolphin (602)	8700	15.4	8.10	10.6	85.8	0.03	0.001
Montezuma Slough at Nurse Slough (609)	4265	14.7	7.97	10.3	46.0	0.00	0.000
Napa River at Vallejo Seawall (340)	23250	14.4	7.92	10.0	22.4	0.00	0.000
Carquinez Strait, west of Benicia army dock (405)	16720	14.5	7.94	10.3	378.3	0.26	0.004

Table B 16-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 4/11/08 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 4/10/08.

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	345	22.3	23.2	7.5	8.8	7.88	8.12	92	64	-
High EC Control @ 17.54 mS	16470	22.4	24.0	6.7	8.8	7.67	7.88	2200	84	-
High EC Control @ 23.43 mS	22570	22.5	23.2	6.6	8.8	7.74	7.85	3000	92	-
Suisun Bay off Chipps Island (508)	5930	22.5	23.5	7.5	8.7	7.96	8.09	710	101	0.002
Grizzly Bay at Dolphin (602)	8300	22.5	23.4	7.2	8.8	7.95	8.05	940	102	0.001
Montezuma Slough at Nurse Slough (609)	4325	22.4	23.3	7.4	8.5	7.95	8.10	540	97	0.000
Napa River at Vallejo Seawall (340)	21650	22.4	23.6	7.2	8.9	7.78	7.92	2640	112	0.000
Carquinez Strait, West of Benicia army dock (405)	15150	22.4	23.3	7.5	8.9	7.85	7.97	1920	108	0.007
DIEPAMHR + 25 ppb PBO	350	22.4	22.5	7.4	8.5	7.91	8.16	-	-	-
High EC Control @ 17.54 mS + 25 ppb PBO	16520	22.4	22.5	7.0	8.4	7.77	7.86	-	-	-
High EC Control @ 23.43 mS + 25 ppb PBO	22010	22.2	23.4	6.7	8.6	7.71	7.83	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	5660	22.1	23.1	7.4	8.7	7.93	8.10	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	8210	22.0	23.2	7.2	8.7	7.95	8.05	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	4152	22.5	23.3	7.6	8.9	7.96	8.08	-	-	-
Napa River at Vallejo Seawall (340) + 25 ppb PBO	21800	22.6	24.0	7.1	8.5	7.79	7.94	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	15180	22.5	24.1	7.3	8.6	7.86	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 B 17-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 4/24/08 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 4/22/08 - 4/23/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	93	7.5	93	4.8	NS
DIEPAMHR + organic matter	100	0.0	95	5.0	NS
Sacramento River at Hood DWR Station	93	2.5	95	2.9	NS
Rough and Ready DWR station, Stockton	98	2.5	98	2.3	NS
Sacramento R. Deep Water Channel, Light 55	97	2.8	98	2.5	NS
Confluence of Linsey Sl. And Cache Sl.	100	0.0	100	0.0	NS
Upper Cache Slough at mouth of Ulatis Creek	98	2.5	98	2.5	NS
Suisun Slough at Rush Ranch	100	0.0	100	0.0	NS
Napa River, near River Park Blvd.	100	0.0	100	0.0	NS
Old River, western arm at railroad bridge (902)	100	0.0	95	5.0	NS
Sacramento River at tip of Grand Island (711)	100	0.0	98	2.3	NS
San Joaquin River at Potato Slough (815)	95	2.9	98	2.5	NS
Old River at mouth of Holland Cut (915)	100	0.0	95	2.9	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.029*	0.005	0.027	0.009	NS
DIEPAMHR + organic matter	0.050	0.005	0.046	0.013	NS
Sacramento River at Hood DWR Station	0.060	0.010	0.042	0.016	NS
Rough and Ready DWR station, Stockton	0.067	0.004	0.051	0.010	NS
Sacramento R. Deep Water Channel, Light 55	0.045	0.005	0.052	0.003	NS
Confluence of Linsey Sl. And Cache Sl.	0.045	0.005	0.038	0.004	NS
Upper Cache Slough at mouth of Ulatis Creek	0.051	0.005	0.062	0.007	NS
Suisun Slough at Rush Ranch	0.042	0.004	0.077	0.002	S*** (183%)

Napa River, near River Park Blvd.	0.046	0.008	0.082	0.005	S** (178%)
Old River, western arm at railroad bridge (902)	0.052	0.004	0.052	0.009	NS
Sacramento River at tip of Grand Island (711)	0.060	0.007	0.057	0.004	NS
San Joaquin River at Potato Slough (815)	0.047	0.011	0.067	0.005	NS
Old River at mouth of Holland Cut (915)	0.054	0.007	0.067	0.004	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

***: $P < 0.001$

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

Table B 17-2. Summary of 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 4/22/08-4/23/08.

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 Hood DWR Station	170	16.8	7.75	9.3	18.8	0.40	0.007
Rough and Ready DWR station, Stockton	579	17.7	7.89	7.9	34.2	0.17	0.004
Sacramento R. Deep Water Channel, Light 55	347	15.9	8.36	10.1	27.0	0.13	0.008
Confluence of Lindsey Sl. And Cache Sl.	297	15.5	8.29	10.2	31.3	0.17	0.008
Upper Cache Slough at mouth of Ulatis Creek	385	15.0	8.40	10.2	46.3	0.04	0.002
Suisun Slough at Rush Ranch	4864	15.1	7.38	7.0	74.5	0.14	0.001
Napa River, near River Park Blvd.	7590	16.6	7.55	8.6	46.3	0.11	0.001
Old River, western arm at railroad bridge (902)	400	15.9	8.22	10.1	15.7	0.02	0.001
Sacramento River at tip of Grand Island (711)	162	16.6	8.15	10.1	9.4	0.53	0.021
San Joaquin River at Potato Slough (815)	305	16.1	8.28	10.1	7.1	0.06	0.003
Old River at mouth of Holland Cut (915)	438	16.4	8.17	10.0	12.5	0.04	0.002

Table B 17-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 4/24/08 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 4/22/08 - 4/23/08.

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	306	21.4	23.7	7.3	8.6	7.85	8.19	100	58	-
DIEPAMHR + organic matter	328	21.4	23.9	7.6	8.3	7.81	8.18	100	58	-
Sacramento River at Hood DWR Station	141	21.4	23.6	7.2	8.8	7.74	7.90	56	62	0.015
Rough and Ready DWR station, Stockton	500	21.3	24.0	7.5	8.6	7.92	8.06	124	88	0.009
Sacramento R. Deep Water Channel, Light 55	295	21.2	24.3	7.3	8.8	8.08	8.20	116	114	0.010
Confluence of Linsey Sl. And Cache Sl.	253	21.2	24.4	7.4	8.8	7.93	8.16	96	108	0.012
Upper Cache Slough at mouth of Ulatis Creek	332	21.2	24.5	7.6	8.8	8.16	8.23	128	132	0.003
Suisun Slough at Rush Ranch	4227	21.3	24.3	7.5	8.5	8.06	8.26	632	192	0.006
Napa River, near River Park Blvd.	6445	21.3	24.3	7.3	8.9	7.85	8.02	860	132	0.003
Old River, western arm at railroad bridge (902)	310	21.3	24.4	7.6	8.8	7.95	8.17	96	86	0.001
Sacramento River at tip of Grand Island (711)	145	21.2	24.4	7.6	8.9	7.77	7.97	72	68	0.024
San Joaquin River at Potato Slough (815)	266	21.2	24.5	7.4	8.9	7.82	8.08	96	88	0.003
Old River at mouth of Holland Cut (915)	356	21.3	24.6	1.8	8.9	7.95	8.08	120	96	0.002
DIEPAMHR + 25 ppb PBO	302	21.3	23.5	7.7	8.6	7.82	8.18	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	331	21.4	23.2	7.6	8.6	7.83	8.27	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	149	21.3	23.4	7.5	8.6	7.75	7.97	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	498	21.4	23.4	7.5	8.6	7.91	8.14	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	296	21.5	23.4	7.5	8.9	8.07	8.16	-	-	-
Confluence of Linsey Sl. And Cache Sl. + 25 ppb PBO	252	21.4	23.4	7.5	8.8	7.95	8.21	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	334	21.4	23.7	7.6	8.8	8.16	8.23	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	4159	21.5	23.5	7.4	8.3	7.87	8.23	-	-	-
Napa River, near River Park Blvd. + 25 ppb PBO	6445	21.4	23.9	7.6	8.7	7.85	8.04	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	300	21.4	23.5	7.4	8.5	7.94	8.15	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	163	21.5	23.8	7.6	8.8	7.75	8.02	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	260	21.5	23.4	7.6	8.9	7.89	8.15	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	362	22.2	23.6	7.6	8.7	7.94	8.13	-	-	-

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 B 18-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 4/25/08 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 4/24/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	95	2.8	97	2.8	NS
DIEPAMHR + organic matter	100	0.0	100	0.0	NS
High EC Control @ 11.20 mS/cm + organic matter	100	0.0	100	0.0	NS
High EC Control @ 22.61 mS/cm + organic matter	100	0.0	80*	5.4	S (80%)
Suisun Bay off Chipps Island (508)	97	2.8	100	0.0	NS
Grizzly Bay at Dolphin (602)	98	2.5	98	2.5	NS
Montezuma Slough at Nurse Slough (609)	98	2.3	98	2.3	NS
Napa River at Vallejo Seawall (340)[4]	98	2.5	90	10.0	NS
Carquinez Strait, West of Benicia army dock (405)[3]	98	2.5	100	0.0	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.017	0.008	0.025	0.003	NS
DIEPAMHR + organic matter	0.049	0.006	0.057	0.004	NS
High EC Control @ 11.20 mS/cm + organic matter	0.060	0.004	0.050	0.006	NS
High EC Control @ 22.61 mS/cm + organic matter	0.021**	0.002	0.048	0.007	S* (229%)
Suisun Bay off Chipps Island (508)	0.061	0.009	0.067	0.006	NS
Grizzly Bay at Dolphin (602)	0.026*	0.008	0.024*	0.010	NS
Montezuma Slough at Nurse Slough (609)	0.070	0.004	0.043*	0.003	S** (61%)
Napa River at Vallejo Seawall (340)[4]	0.023	0.007	0.050	0.009	NS
Carquinez Strait, West of Benicia army dock (405)[3]	0.038	0.012	0.028*	0.004	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

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

3. This high conductivity sample was compared to the High EC control @ 11.20 mS/cm.

4. This high conductivity sample was compared to the High EC control @ 22.60 mS/cm.

Table B 18-2. Summary of 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 4/25/08.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (^oC)	pH	DO (mg/L)			
Suisun Bay off Chipps Island (508)	2207	14.9	8.10	10.5	25.7	0.12	0.003
Grizzly Bay at Dolphin (602)	6520	15.0	8.10	10.6	33.7	0.10	0.003
Montezuma Slough at Nurse Slough (609)	4609	14.2	7.80	9.5	60.7	0.14	0.002
Napa River at Vallejo Seawall (340)	22080	15.1	7.92	10.2	18.5	0.09	0.001
Carquinez Strait, West of Benicia army dock (405)	10360	14.1	8.00	10.4	141.7	0.20	0.004

Table B 18-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 4/25/08 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 4/24/08.

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 (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	314	22.2	25.0	7.5	8.4	7.84	8.05	100	58	-
DIEPAMHR + organic matter	342	22.3	24.8	7.5	8.3	7.73	8.04	100	58	-
High EC Control @ 11.20 mS/cm + organic matter	10010	22.9	25.0	7.3	8.7	7.66	7.80	1260	84	-
High EC Control @ 22.61 mS/cm + organic matter	20000	22.8	24.9	5.7	8.3	7.57	7.78	3280	90	-
Suisun Bay off Chipps Island (508)	1801	21.9	24.9	7.5	8.7	7.93	8.06	244	92	0.006
Grizzly Bay at Dolphin (602)	6015	22.5	25.1	7.5	8.9	7.90	8.00	748	102	0.003
Montezuma Slough at Nurse Slough (609)	4183	22.3	25.0	7.5	8.5	7.84	9.17	576	102	0.004
Napa River at Vallejo Seawall (340)	19395	21.5	24.8	7.0	8.4	7.77	7.89	2368	116	0.002
Carquinez Strait, West of Benicia army dock (405)	9180	22.5	24.7	7.2	8.8	7.88	7.95	1144	102	0.006
DIEPAMHR + 25 ppb PBO	310	21.8	24.1	7.3	8.5	7.77	8.02	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	379	22.6	23.7	7.3	8.4	7.80	8.01	-	-	-
High EC Control @ 11.20 mS/cm + 25 ppb PBO	9760	22.3	24.0	7.2	8.6	7.65	7.78	-	-	-
High EC Control @ 22.61 mS/cm + 25 ppb PBO	19595	23.3	24.0	6.7	8.3	7.65	7.79	-	-	-

Suisun Bay off Chipps Island (508) + 25 ppb PBO	1825	23.2	24.0	7.4	8.5	7.94	8.01	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	5955	22.6	23.6	7.5	8.6	7.92	7.99	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	4173	23.6	24.0	7.3	8.7	7.88	7.96	-	-	-
Napa River at Vallejo Seawall (340) + 25 ppb PBO	19300	23.0	23.7	6.2	8.3	7.76	7.89	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	8855	23.3	23.9	7.4	8.8	7.86	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 B 19-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 5/08/08 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 5/06/08 - 5/07/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	95	2.9	98	2.5	NS
DIEPAMHR + organic matter	100	0.0	97	2.8	NS
High EC Control @10.83 mS + organic matter	100	0.0	93	4.8	NS
High EC Control @17.36 mS + organic matter	98	2.5	98	2.5	NS
Suisun Slough at Rush Ranch	100	0.0	100	0.0	NS
Napa River at River Park Blvd.	100	0.0	98	2.5	NS
Montezuma Slough at Nurse Slough (609)	95	2.9	100	0.0	NS
Grizzly Bay at Dolphin (602)[3]	100	0.0	98	2.3	NS
Suisun Bay off Chipps Island (508)	100	0.0	100	0.0	NS
Carquinez Strait, West of Benicia army dock (405)[4]	100	0.0	100	0.0	NS
Trip Blank: DIEPAMHR	98	2.5	-	-	NA
Field Dup.: Montezuma Slough at Nurse Slough (609)	100	0.0	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.042*	0.006	0.040**	0.006	NS
DIEPAMHR + organic matter	0.059	0.006	0.070	0.006	NS
High EC Control @10.83 mS + organic matter	0.081	0.009	0.066	0.003	NS
High EC Control @17.36 mS + organic matter	0.081	0.006	0.043*	0.011	S* (53%)
Suisun Slough at Rush Ranch	0.082	0.006	0.067	0.005	NS
Napa River at River Park Blvd.	0.087	0.003	0.071	0.009	NS

Montezuma Slough at Nurse Slough (609)	0.087	0.006	0.077	0.006	NS
Grizzly Bay at Dolphin (602)[3]	0.062*	0.004	0.044***	0.003	S* (71%)
Suisun Bay off Chipps Island (508)	0.103	0.004	0.071	0.006	S** (69%)
Carquinez Strait, West of Benicia army dock (405)[4]	0.050**	0.001	0.043	0.004	NS
Trip Blank: DIEPAMHR	0.045	0.006	-	-	NA
Field Dup.: Montezuma Slough at Nurse Slough (609)	0.074	0.010	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

   *: $P < 0.05$

   **: $P < 0.01$

   ***: $P < 0.001$

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

3. This high conductivity sample was compared to the High EC Control @ 10.83 mS/cm.

4. This high conductivity sample was compared to the High EC Control @ 17.36 mS/cm.

Table B 19-2. Summary of 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 5/06/08 - 5/07/08.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Suisun Slough at Rush Ranch	5740	15.4	7.81	7.6	154.5	0.20	0.003
Napa River at River Park Blvd.	8430	18.3	8.10	8.2	58	0.04	0.002
Montezuma Slough at Nurse Slough (609)	5030	17.6	7.74	8.8	59.4	0.08	0.001
Grizzly Bay at Dolphin (602)	11260	17.0	8.01	9.5	213.5	0.15	0.003
Suisun Bay off Chipps Island (508)	5310	16.9	7.80	9.7	38.3	0.09	0.001
Carquinez Strait, West of Benicia army dock (405)	17080	14.9	7.86	10.4	252.5	0.21	0.003
Trip Blank: DIEPAMHR	345	21.6	8.15	8.6	0.25	0.02	0.001
Field Dup.: Montezuma Slough at Nurse Slough (609)	5030	17.6	7.74	8.8	55.9	0.09	0.001

Table B 19-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 5/08/08 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 5/06/08 - 5/07/08.

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	334	24.2	24.6	7.2	8.1	7.61	8.10	88	58	-
DIEPAMHR + organic matter	330	24.0	24.3	7.0	8.2	7.61	8.15	88	58	-
High EC Control @10.83 mS + organic matter	10205	23.9	24.3	7.1	8.4	7.60	7.88	1640	74	-
High EC Control @17.36 mS + organic matter	16450	23.7	24.3	6.9	8.2	7.57	7.90	2600	82	-
Suisun Slough at Rush Ranch	5300	24.0	24.3	7.1	8.6	7.91	8.14	720	156	0.006
Napa River at River Park Blvd.	7980	23.9	24.2	7.1	8.9	7.84	8.05	960	140	0.001
Montezuma Slough at Nurse Slough (609)	4653	24.2	24.3	7.2	8.7	7.81	7.95	680	110	0.003
Grizzly Bay at Dolphin (602)	9660	24.2	24.2	7.1	8.7	7.80	7.95	1560	102	0.005
Suisun Bay off Chipps Island (508)	4870	23.9	24.2	7.3	8.5	7.82	7.92	660	94	0.003
Carquinez Strait, West of Benicia army dock (405)	15380	24.2	24.2	7.0	8.2	7.71	7.83	2480	104	0.005
Trip Blank: DIEPAMHR	478	24.2	24.6	7.4	8.6	7.82	8.18	120	60	0.001
Field Dup.: Montezuma Slough at Nurse Slough (609)	4763	24.2	24.2	7.3	8.6	7.89	8.09	740	108	0.004
DIEPAMHR + 25 ppb PBO	401	23.7	24.2	7.4	8.3	7.71	8.14	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	334	23.5	24.2	7.2	8.1	7.60	8.16	-	-	-
High EC Control @10.83 mS + organic matter + 25 ppb PBO	10080	24.0	24.2	7.2	8.2	7.55	7.96	-	-	-
High EC Control @17.36 mS + organic matter + 25 ppb PBO	16385	24.0	24.2	7.0	8.2	7.53	7.93	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	5309	24.1	24.1	7.3	8.3	7.96	8.12	-	-	-
Napa River at River Park Blvd. + 25 ppb PBO	7970	24.2	24.4	7.2	8.3	7.92	8.04	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	4692	24.1	24.4	7.2	8.4	7.94	7.95	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	9740	24.2	24.3	7.1	8.3	7.05	8.01	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	5125	24.2	24.3	7.3	8.7	7.84	8.08	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	15475	23.9	24.3	7.0	8.2	7.77	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 B 20-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 5/09/08 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 5/08/08 - 5/09/08.

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	90	4.1	NS
Low EC Control + organic matter	98	2.5	95	2.9	NS
Upper Cache Slough at mouth of Ulatis Creek	100	0.0	100	0.0	NS
Sacramento River at tip of Grand Island (711)[3]	100	0.0	93	4.8	NS
Confluence of Lindsey Sl. And Cache Sl.	100	0.0	100	0.0	NS
Sacramento R. Deep Water Channel, Light 55	100	0.0	100	0.0	NS
Rough and Ready DWR station, Stockton	100	0.0	100	0.0	NS
Sacramento River at Hood DWR Station[3]	98	2.5	100	0.0	NS
Bottle Blank	100	0.0	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.012***	0.004	0.033**	0.003	S** (275%)
DIEPAMHR + organic matter	0.082	0.007	0.076	0.010	NS
Low EC Control + organic matter	0.058*	0.004	0.074	0.011	NS
Upper Cache Slough at mouth of Ulatis Creek	0.048**	0.006	0.067	0.005	NS
Sacramento River at tip of Grand Island (711)[3]	0.037*	0.009	0.063	0.013	NS
Confluence of Lindsey Sl. And Cache Sl.	0.055*	0.006	0.073	0.006	NS
Sacramento R. Deep Water Channel, Light 55	0.063	0.011	0.076	0.005	NS
Rough and Ready DWR station, Stockton	0.064	0.007	0.084	0.006	NS
Sacramento River at Hood DWR Station[3]	0.043	0.011	0.061	0.010	NS
Bottle Blank	0.036	0.007	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

***: $P < 0.001$

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

3. These low conductivity sample was compared to the Low EC Control.

Table B 20-2. Summary of 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 5/08/08 - 5/09/08.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Upper Cache Slough at mouth of Ulatis Creek	319	17.4	8.13	8.8	58.8	0.15	0.006
Sacramento River at tip of Grand Island (711)	181	18.8	7.72	11.7	14.1	0.50	0.009
Confluence of Lindsey Sl. And Cache Sl.	245	17.8	7.96	8.8	31.4	0.31	0.009
Sacramento R. Deep Water Channel, Light 55	251	17.9	7.85	8.9	31.5	0.23	0.005
Rough and Ready DWR station, Stockton	410	20.1	7.86	7.8	9.0	0.05	0.001
Sacramento River at Hood DWR Station	192	20.8	7.81	8.0	6.4	0.34	0.008

Table B 20-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 5/09/08 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 5/08/08 - 5/09/08.

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	302	22.9	23.5	7.3	8.1	7.72	8.18	104	60	-
DIEPAMHR + organic matter	355	23.1	24.4	7.0	8.3	7.62	8.16	104	60	-
Low EC Control + organic matter	197	23.1	24.3	7.1	8.2	7.34	8.01	68	28	-
Upper Cache Slough at mouth of Ulatis Creek	287	23.0	24.6	7.2	8.5	7.97	8.30	112	106	0.007
Sacramento River at tip of Grand Island (711)	171	23.0	24.0	7.0	8.5	7.68	8.10	64	72	0.030
Confluence of Lindsey Sl. And Cache Sl.	223	23.0	24.6	7.0	8.7	7.82	8.16	84	84	0.015
Sacramento R. Deep Water Channel, Light 55	252	23.0	24.1	7.1	8.6	7.83	8.24	96	94	0.007
Rough and Ready DWR station, Stockton	378	23.0	24.1	7.2	8.6	7.67	7.91	96	66	0.003
Sacramento River at Hood DWR Station	177	23.0	24.1	7.0	8.5	7.67	7.78	80	72	0.016
Bottle Blank	329	23.0	24.6	7.4	8.9	7.77	8.19	-	-	-
DIEPAMHR + 25 ppb PBO	322	23.0	23.4	7.3	8.2	7.71	8.15	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	351	23.0	23.3	7.0	8.3	7.63	8.17	-	-	-
Low EC Control + organic matter + 25 ppb PBO	205	23.0	23.5	7.1	8.7	7.43	7.95	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	285	23.0	23.8	7.3	8.6	7.98	8.22	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	171	23.0	23.9	7.0	8.7	7.65	8.22	-	-	-
Confluence of Lindsey Sl. And Cache Sl. + 25 ppb PBO	220	23.1	23.4	6.8	8.7	7.82	8.28	-	-	-

Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	250	23.1	24.1	7.0	8.6	7.83	8.26	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	382	23.1	23.5	7.2	8.7	7.70	7.81	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	186	23.1	23.7	7.1	8.5	7.65	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 B 21-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 5/13/08 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 5/12/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	98	2.5	95	2.8	NS
DIEPAMHR + organic matter	92	2.6	100	0.0	NS
Old River, western arm at railroad bridge (902)	100	0.0	98	2.5	NS
Old River at mouth of Holland Cut (915)	98	2.5	100	0.0	NS
San Joaquin River at Potato Slough (815)	100	0.0	100	0.0	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.044**	0.005	0.034**	0.009	NS
DIEPAMHR + organic matter	0.088	0.011	0.087	0.009	NS
Old River, western arm at railroad bridge (902)	0.097	0.006	0.091	0.020	NS
Old River at mouth of Holland Cut (915)	0.090	0.008	0.090	0.006	NS
San Joaquin River at Potato Slough (815)	0.071	0.003	0.088	0.004	S* (124%)

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

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

Table B 21-2. Summary of 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 5/12/08.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)[1]
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Old River, western arm at railroad bridge (902)	348	18.5	7.74	8.8	21.2	0.02	0.000
Old River at mouth of Holland Cut (915)	471	19.8	7.69	8.5	10.5	0.04	0.001
San Joaquin River at Potato Slough (815)	321	18.8	7.78	9.1	8.4	0.12	0.002

1. Unionized ammonia was calculated from ammonia nitrogen measured at sample receipt and water chemistry measured at sample collection.

Table B 21-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 5/13/08 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 5/12/08.

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	22.9	24.0	7.3	8.5	7.77	8.03	88	58	-
DIEPAMHR + organic matter	340	22.6	24.5	7.2	8.5	7.64	7.91	88	58	-
Old River, western arm at railroad bridge (902)	316	23.2	24.1	7.4	8.6	7.86	8.01	88	82	0.001
Old River at mouth of Holland Cut (915)	418	22.5	23.8	7.3	8.3	7.86	8.07	108	88	0.001
San Joaquin River at Potato Slough (815)	286	23.1	24.0	7.4	8.6	7.82	8.03	84	76	0.005
DIEPAMHR + 25 ppb PBO	341	23.5	24.0	7.4	8.3	7.78	8.01	-	-	-

DIEPAMHR + organic matter + 25 ppb PBO	326	23.6	23.8	7.3	8.7	7.65	7.96	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	330	23.8	24.1	7.3	8.6	7.87	8.02	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	418	23.4	23.8	7.4	8.5	7.89	8.06	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	293	23.8	24.5	7.3	8.7	7.85	8.05	-	-	-

1. Unionized ammonia was calculated from ammonia nitrogen measured at sample receipt and water chemistry measured at test initiation.

Table B 22-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 5/22/08 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 5/20/08 - 5/21/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	98	2.5	98	2.5	NS
DIEPAMHR + organic matter	98	2.5	95	2.8	NS
Low EC Control + organic matter	98	2.5	95	3.1	NS
High EC Control + organic matter	97	2.8	100	0.0	NS
Sacramento River at Hood DWR Station[3]	100	0.0	100	0.0	NS
Rough and Ready DWR station, Stockton	100	0.0	100	0.0	NS
Napa River at River Park Blvd.[4]	100	0.0	98	2.5	NS
Suisun Slough at Rush Ranch	100	0.0	98	2.5	NS
Confluence of Linsey Sl. And Cache Sl.	100	0.0	98	2.5	NS
Upper Cache Slough at mouth of Ulatis Creek	100	0.0	100	0.0	NS
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

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.084	0.007	0.080	0.003	NS
DIEPAMHR + organic matter	0.110	0.008	0.081	0.003	S* (74%)
Low EC Control + organic matter	0.112	0.010	0.096	0.007	NS
High EC Control + organic matter	0.063	0.017	0.085	0.006	NS
Sacramento River at Hood DWR Station[3]	0.114	0.003	0.094	0.004	S* (82%)
Rough and Ready DWR station, Stockton	0.124	0.017	0.101	0.009	NS
Napa River at River Park Blvd.[4]	0.116	0.007	0.055**	0.004	S*** (47%)
Suisun Slough at Rush Ranch	0.116	0.009	0.098	0.007	NS

Confluence of Linsey Sl. And Cache Sl.	0.093	0.011	0.119	0.013	NS
Upper Cache Slough at mouth of Ulatis Creek	0.105	0.001	0.093	0.008	NS
Sacramento R. Deep Water Channel, Light 55	0.107	0.005	0.108	0.004	NS
Sacramento River at tip of Grand Island (711)[3]	0.116	0.005	0.098	0.006	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

   *: $P < 0.05$

   **: $P < 0.01$

   ***: $P < 0.001$

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

3. These low conductivity samples were compared to the Low EC Control.

4. This high conductivity sample was compared to the High EC Control.

Table B 22-2. Summary of 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 5/20/08 - 5/21/08.

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 Hood DWR Station	180	24.1	7.32	7.9	16.7	0.41	0.004
Rough and Ready DWR station, Stockton	401	23.1	7.08	7.3	11.7	0.07	0.000
Napa River at River Park Blvd.	12540	22.6	7.51	5.9	24.3	0.06	0.001
Suisun Slough at Rush Ranch	6870	16.9	7.38	6.7	46.8	0.11	0.001
Confluence of Lindsey Sl. And Cache Sl.	243	19.5	7.22	8.7	72.1	0.23	0.001
Upper Cache Slough at mouth of Ulatis Creek	288	19.2	7.19	9.2	94.3	0.14	0.001
Sacramento R. Deep Water Channel, Light 55	280	20.7	7.47	8.8	40.2	0.17	0.002
Sacramento River at tip of Grand Island (711)	179	22.9	6.93	8.6	31.6	0.43	0.002

Table B 22-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 5/22/08 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 5/20/08 - 5/21/08.

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 (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	346	23.2	23.9	7.4	8.2	7.36	7.82	104	60	-
DIEPAMHR + organic matter	347	23.2	23.9	6.8	8.2	7.44	7.80	104	60	-
Low EC Control + organic matter	184	23.2	24.1	7.1	8.7	7.45	7.78	54	35	-
High EC Control + organic matter	12145	23.2	24.4	7.0	8.8	7.31	7.80	1400	79	-
Sacramento River at Hood DWR Station	192	23.2	24.6	6.6	8.7	7.56	7.90	64	66	0.008
Rough and Ready DWR station, Stockton	398	23.1	24.5	6.9	8.9	7.57	7.98	88	64	0.001
Napa River at River Park Blvd.	12330	23.1	24.5	6.9	8.7	7.05	8.11	1400	148	0.000
Suisun Slough at Rush Ranch	6800	23.1	24.5	7.3	8.8	7.12	8.20	800	168	0.001
Confluence of Linsey Sl. And Cache Sl.	304	22.9	24.4	6.9	8.4	7.68	8.16	80	88	0.005
Upper Cache Slough at mouth of Ulatis Creek	274	23.0	24.4	6.9	8.3	7.70	8.11	96	102	0.003
Sacramento R. Deep Water Channel, Light 55	288	23.0	24.3	7.0	8.7	7.69	8.08	88	92	0.004
Sacramento River at tip of Grand Island (711)	185	22.8	24.0	7.1	8.4	7.63	7.97	64	68	0.009
DIEPAMHR + 25 ppb PBO	347	22.8	24.1	7.3	8.3	7.62	7.85	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	347	22.8	23.9	7.0	8.4	7.61	7.91	-	-	-
Low EC Control + organic matter + 25 ppb PBO	6149	22.8	23.9	7.2	8.8	7.24	7.69	-	-	-
High EC Control + organic matter + 25 ppb PBO	6171	22.8	23.6	7.2	8.7	7.62	7.85	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	198	22.9	23.7	6.9	8.2	7.52	7.90	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	395	22.8	24.1	7.2	8.4	7.66	7.96	-	-	-
Napa River at River Park Blvd. + 25 ppb PBO	12045	22.7	22.9	7.1	8.5	7.08	8.10	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	6630	22.7	22.9	7.3	8.7	7.12	8.19	-	-	-
Confluence of Linsey Sl. And Cache Sl. + 25 ppb PBO	281	22.8	23.1	7.1	8.8	7.64	8.17	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	292	22.7	23.3	7.1	8.6	7.85	8.12	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	287	22.9	23.2	6.8	8.7	7.73	8.07	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	186	22.8	23.5	7.0	8.4	7.65	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 B 23-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 5/25/08 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 5/23/08.

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	95	2.9	93	2.5	NS
High EC Control @ 13.77 mS + organic matter	100	0.0	95	2.9	NS
High EC Control @ 18.91 mS + organic matter	100	0.0	100	0.0	NS
Suisun Bay off Chipps Island (508)	100	0.0	100	0.0	NS
San Joaquin River at Potato Slough (815)	98	2.5	100	0.0	NS
Old River, western arm at railroad bridge (902)	100	0.0	97	2.8	NS
Carquinez Strait, West of Benicia army dock (405)[4]	98	2.5	94	3.7	NS
Old River at mouth of Holland Cut (915)	100	0.0	100	0.0	NS
Grizzly Bay at Dolphin (602)[3]	95	3.2	95	2.9	NS
Montezuma Slough at Nurse Slough (609)	100	0.0	100	0.0	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.037**	0.014	0.025	0.010	NS
DIEPAMHR + organic matter	0.060	0.003	0.061	0.010	NS
High EC Control @ 13.77 mS + organic matter	0.040	0.005	0.054	0.014	NS
High EC Control @ 18.91 mS + organic matter	0.027*	0.012	0.048*	0.007	NS
Suisun Bay off Chipps Island (508)	0.066	0.008	0.091	0.002	S* (138%)
San Joaquin River at Potato Slough (815)	0.076	0.003	0.110	0.004	S*** (145%)
Old River, western arm at railroad bridge (902)	0.053	0.016	0.098	0.005	S* (185%)
Carquinez Strait, West of Benicia army dock (405)[4]	0.057	0.008	0.024	0.006	S* (42%)
Old River at mouth of Holland Cut (915)	0.081	0.007	0.094	0.004	NS
Grizzly Bay at Dolphin (602)[3]	0.033	0.010	0.061	0.004	S* (185%)
Montezuma Slough at Nurse Slough (609)	0.056	0.005	0.073	0.006	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

***: $P < 0.001$

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

3. This high conductivity sample was compared to the High EC Control @ 13.77 mS/cm

4. This high conductivity sample was compared to the High EC Control @ 18.91 mS/cm

Table B 23-2. Summary of 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 5/23/08.

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)	6200	18.7	7.15	9.3	18.7	0.06	0.002
San Joaquin River at Potato Slough (815)	317	19.8	7.21	9.6	12.0	0.06	0.003
Old River, western arm at railroad bridge (902)	359	19.1	6.59	8.8	42.8	0.51	0.020
Carquinez Strait, West of Benicia army dock (405)	18670	17.9	7.49	10.1	225.3	0.19	0.005
Old River at mouth of Holland Cut (915)	446	20.6	7.23	9.4	8.9	0.05	0.003
Grizzly Bay at Dolphin (602)	13290	18.4	7.64	10.0	82.7	0.10	0.003
Montezuma Slough at Nurse Slough (609)	7060	19.1	7.28	9.2	50.9	0.09	0.002

Table B 23-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 5/25/08 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 5/23/08.

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 (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	323	20.4	23.4	7.8	8.6	7.79	8.13	104	62	-
DIEPAMHR + organic matter	323	20.9	23.1	7.5	8.4	7.68	8.08	104	62	-
High EC Control @ 13.77 mS + organic matter	12536	20.9	23.6	7.5	8.3	7.59	8.01	1540	83	-
High EC Control @ 18.91 mS + organic matter	17575	20.9	23.4	7.3	8.3	7.61	8.02	2220	94	-
Suisun Bay off Chipps Island (508)	5805	21.3	23.5	7.6	8.7	7.79	8.07	668	94	0.002
San Joaquin River at Potato Slough (815)	320	22.0	23.4	7.6	8.8	7.93	8.19	84	80	0.003
Old River, western arm at railroad bridge (902)	335	21.9	23.6	7.6	8.7	7.89	8.14	112	84	0.020
Carquinez Strait, West of Benicia army dock (405)	17855	22.1	23.8	7.6	8.8	7.80	7.99	224	112	0.005
Old River at mouth of Holland Cut (915)	415	22.0	23.4	7.5	8.4	8.01	8.27	116	86	0.003
Grizzly Bay at Dolphin (602)	12665	22.0	23.1	7.5	8.5	7.84	8.01	148	106	0.003
Montezuma Slough at Nurse Slough (609)	6490	21.7	23.2	7.5	8.8	7.87	8.13	792	112	0.002
DIEPAMHR + 25 ppb PBO	360	21.8	23.2	7.5	8.5	7.81	8.14	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	346	22.1	22.4	7.6	8.5	7.77	8.11	-	-	-
High EC Control @ 13.77 mS + organic matter + 25 ppb PBO	12620	21.7	23.1	7.3	8.6	7.67	8.01	-	-	-
High EC Control @ 18.91 mS + organic matter + 25 ppb PBO	17550	21.9	23.1	7.3	8.2	7.69	8.03	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	5745	22.1	22.9	7.4	8.7	7.80	8.07	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	310	21.8	23.2	7.5	8.9	7.87	8.14	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	348	22.0	22.5	7.4	8.5	7.92	8.12	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	16970	21.8	22.7	7.5	8.1	7.81	7.95	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	411	21.9	22.1	7.5	8.7	7.87	8.21	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	12115	21.9	22.8	7.5	8.3	7.83	8.02	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	6520	21.9	22.5	7.5	8.5	7.89	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 B 24-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 6/5/08 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 6/03/08 - 6/04/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	95	2.9	98	2.5	NS
DIEPAMHR + POM	94	3.3	93	4.8	NS
Low EC Control @ 210 uS + organic matter	98	2.5	90	4.1	NS
High EC Control @ 12.32 mS + organic matter	100	0.0	98	2.5	NS
High EC Control @ 17.95 mS + organic matter	95	2.8	93	7.5	NS
Sacramento River at Hood DWR Station[3]	100	0.0	100	0.0	NS
Rough and Ready DWR station, Stockton	98	2.5	100	0.0	NS
Napa River at River Park Blvd.[5]	100	0.0	93	4.4	NS
Suisun Slough at Rush Ranch	100	0.0	98	2.3	NS
Montezuma Slough at Nurse Slough (609)	100	0.0	100	0.0	NS
Grizzly Bay at Dolphin (602)[4]	100	0.0	95	4.5	NS
Carquinez Strait, West of Benicia army dock (405)[4]	100	0.0	95	3.1	NS
Suisun Bay off Chipps Island (508)[4]	98	2.5	98	2.5	NS
Field Dup.: Montezuma Slough at Nurse Slough (609)	100	0.0	-	-	NA
Trip Blank: DIEPAMHR	100	0.0	-	-	NA
Bottle Blank: DIEPAMHR	95	2.9	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.053	0.003	0.045*	0.007	NS
DIEPAMHR + organic matter	0.062	0.007	0.068	0.005	NS
Low EC Control @ 210 uS + organic matter	0.098	0.007	0.092	0.005	NS
High EC Control @ 12.32 mS + organic matter	0.049	0.004	0.045**	0.003	NS
High EC Control @ 17.95 mS + organic matter	0.028**	0.002	0.032***	0.003	NS
Sacramento River at Hood DWR Station[3,6]	0.092	0.007	0.076*	0.002	NS
Rough and Ready DWR station, Stockton	0.093	0.010	0.105	0.011	NS
Napa River at River Park Blvd.[5]	0.087	0.028	0.049	0.006	NS
Suisun Slough at Rush Ranch	0.092	0.006	0.089	0.004	NS
Montezuma Slough at Nurse Slough (609)	0.089	0.007	0.088	0.011	NS
Grizzly Bay at Dolphin (602)[4]	0.069	0.004	0.048	0.007	S* (70%)
Carquinez Strait, West of Benicia army dock (405)[4]	0.059	0.004	0.052	0.008	NS
Suisun Bay off Chipps Island (508)[4]	0.101	0.008	0.078	0.004	S* (77%)
Field Dup.: Montezuma Slough at Nurse Slough (609)	0.102	0.006	-	-	NA
Trip Blank: DIEPAMHR	0.047	0.004	-	-	NA
Bottle Blank: DIEPAMHR	0.046	0.009	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

***: $P < 0.001$

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

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

4. This high conductivity sample was compared to the High EC Control @ 12.32 mS/cm.

5. This high conductivity sample was compared to the High EC Control @ 17.95 mS/cm.

6. The PBO-treated Hood sample showed significantly lower weight compared to the Low EC Control, but not compared to the DIEPAMHR or DIEPAMHR + POM.

Table B 24-2. Summary of 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 6/03/08 - 6/04/08.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (^{o}C)	pH	DO (mg/L)			
Sacramento River at Hood DWR Station	214	20.8	7.23	8.2	17.5	0.30	0.007
Rough and Ready DWR station, Stockton	498	20.8	7.28	6.8	18.2	0.04	0.001
Napa River at River Park Blvd.	17040	20.0	7.49	7.3	30.0	0.00	0.000
Suisun Slough at Rush Ranch	8580	16.7	6.74	6.3	101.7	0.09	0.002
Montezuma Slough at Nurse Slough (609)	8160	19.1	7.56	9.1	50.8	0.12	0.003
Grizzly Bay at Dolphin (602)	11930	18.8	7.39	9.4	104.0	0.18	0.007
Carquinez Strait, West of Benicia army dock (405)	18600	17.7	7.86	9.5	95.4	0.19	0.004
Suisun Bay off Chipps Island (508)	5450	18.0	7.30	9.7	33.3	0.14	0.005
Field Dup.: Montezuma Slough at Nurse Slough (609)	8160	19.1	7.56	9.1	56.7	0.14	0.001
Trip Blank: DIEPAMHR	330	21.3	8.10	8.8	0.2	0.00	0.000
Bottle Blank: DIEPAMHR	-	-	-	-	0.3	-	-

Table B 24-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 6/05/08 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 6/03/08 - 6/04/08.

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	22.6	23.9	7.4	8.2	7.66	8.17	100	60	-
DIEPAMHR + organic matter	350	22.6	23.6	7.3	8.3	7.63	8.18	100	60	-
Low EC Control @ 210 uS + organic matter	222	22.6	23.3	7.1	8.6	7.37	7.99	60	34	-
High EC Control @ 12.32 mS + organic matter	11945	22.6	23.4	7.1	8.5	7.67	7.92	1320	74	-
High EC Control @ 17.95 mS + organic matter	17400	22.6	23.3	7.1	8.7	7.70	7.93	1920	80	-
Sacramento River at Hood DWR Station	207	22.6	24.2	7.1	8.4	7.72	7.90	60	66	0.009
Rough and Ready DWR station, Stockton	488	22.6	23.7	7.2	8.8	7.79	8.02	124	74	0.002
Napa River at River Park Blvd.	16785	22.6	23.3	6.6	8.5	7.74	8.02	1920	140	0.000
Suisun Slough at Rush Ranch	8300	22.6	23.8	7.1	8.8	7.45	8.15	960	148	0.001
Montezuma Slough at Nurse Slough (609)	7745	22.6	23.6	7.4	8.4	7.91	8.04	880	112	0.004
Grizzly Bay at Dolphin (602)	11710	22.6	24.1	7.0	8.6	7.79	7.90	1280	92	0.005
Carquinez Strait, West of Benicia army dock (405)	11635	22.6	23.7	7.2	8.4	7.75	7.98	1880	100	0.007
Suisun Bay off Chipps Island (508)	11110	22.6	23.0	7.2	8.3	7.81	7.94	60	86	0.003
Field Dup.: Montezuma Slough at Nurse Slough (609)	7800	22.6	24.1	7.3	8.7	7.88	8.03	840	108	0.004
Trip Blank: DIEPAMHR	378	22.6	24.2	7.3	8.9	7.87	8.19	112	58	0.000
Bottle Blank: DIEPAMHR	407	22.9	23.7	7.4	8.6	7.81	8.25	-	-	-
DIEPAMHR + 25 ppb PBO	383	22.6	23.6	7.4	8.2	7.74	8.19	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	372	22.7	23.8	7.2	8.4	7.62	8.16	-	-	-
Low EC Control @ 210 uS + organic matter + 25 ppb PBO	242	22.7	23.8	7.3	8.6	7.35	7.96	-	-	-
High EC Control @ 12.32 mS + organic matter + 25 ppb PBO	11990	22.7	24.2	7.2	8.6	7.67	7.92	-	-	-
High EC Control @ 17.95 mS + organic matter + 25 ppb PBO	17325	22.6	23.8	7.1	8.5	7.70	7.93	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	238	22.7	23.6	7.2	8.5	7.74	7.98	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	509	22.7	23.7	7.1	8.5	7.82	8.10	-	-	-
Napa River at River Park Blvd. + 25 ppb PBO	16835	22.7	23.9	6.8	8.1	7.76	8.02	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	8240	22.7	23.7	7.1	8.5	7.85	8.15	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	7690	22.9	23.6	7.3	8.6	7.85	8.03	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	11240	23.3	23.3	7.1	8.5	7.70	7.91	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	17380	23.0	23.4	7.1	8.4	7.78	7.89	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	5195	22.9	23.5	7.2	8.7	7.83	7.96	-	-	-

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 B 25-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 6/6/08 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 6/6/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	95	2.9	96	4.2	NS
DIEPAMHR + organic matter	98	2.5	94	3.2	NS
Low EC Control @ 200 uS + organic matter	75*	9.0	87	2.3	NS
Sacramento River at tip of Grand Island (711)	100	0.0	100	0.0	NS
Upper Cache Slough at mouth of Ulatis Creek	100	0.0	98	2.5	NS
Confluence of Lindsey Sl. And Cache Sl.	98	2.5	100	0.0	NS
Sacramento R. Deep Water Channel, Light 55	98	2.5	97	2.8	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.037**	0.007	0.045	0.008	NS
DIEPAMHR + organic matter	0.079	0.007	0.062	0.005	NS
Low EC Control @ 200 uS + organic matter	0.050*	0.007	0.067	0.010	NS
Sacramento River at tip of Grand Island (711)	0.081	0.007	0.107	0.004	NS
Upper Cache Slough at mouth of Ulatis Creek	0.084	0.010	0.077	0.014	NS
Confluence of Lindsey Sl. And Cache Sl.	0.097	0.013	0.091	0.001	NS
Sacramento R. Deep Water Channel, Light 55	0.086	0.014	0.097	0.005	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

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

Table B 25-2. Summary of 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 6/6/08.

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)	217	18.8	7.07	8.8	20.2	0.24	0.001
Upper Cache Slough at mouth of Ulatis Creek	403	17.9	7.35	8.7	92.2	0.20	0.001
Confluence of Lindsey Sl. And Cache Sl.	351	18.9	7.16	9.4	48.5	0.16	0.001
Sacramento R. Deep Water Channel, Light 55	436	19.7	7.35	8.8	63.9	0.15	0.001

Table B 25-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 6/6/08 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 6/6/08.

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	23.0	23.6	7.5	8.2	7.76	8.11	100	60	-
DIEPAMHR + organic matter	347	23.0	23.0	7.2	8.5	7.65	8.06	100	60	-
Low EC Control @ 200 uS + organic matter	207	23.0	23.6	7.1	8.9	7.48	7.92	56	30	-
Sacramento River at tip of Grand Island (711)	208	23.0	23.6	7.2	8.9	7.66	7.94	68	70	0.010
Upper Cache Slough at mouth of Ulatis Creek	265	23.1	24.0	7.4	8.8	7.79	7.97	44	90	0.009
Confluence of Lindsey Sl. And Cache Sl.	220	23.1	24.3	7.3	8.8	7.73	7.90	68	78	0.006
Sacramento R. Deep Water Channel, Light 55	261	23.3	23.3	7.2	8.9	7.75	7.95	84	82	0.006
DIEPAMHR + 25 ppb PBO	340	22.9	23.2	7.3	8.4	7.70	8.03	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	341	23.2	23.4	7.1	8.4	7.60	8.03	-	-	-
Low EC Control @ 200 uS + organic matter + 25 ppb PBO	202	23.5	23.7	7.0	8.8	7.37	7.95	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	211	23.4	24.0	7.3	8.9	7.63	7.90	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	256	23.8	24.0	7.2	8.7	7.85	7.97	-	-	-
Confluence of Lindsey Sl. And Cache Sl. + 25 ppb	207	23.4	23.6	7.4	8.8	7.74	7.88	-	-	-

PBO										
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	265	23.8	24.0	7.4	8.7	7.78	7.96	-	-	-

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 B 26-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 6/19/08 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 6/17/08 - 6/18/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	97	2.8	94	3.3	NS
DIEPAMHR + organic matter	98	2.5	95	3.1	NS
Low EC Control @ 139.6 uS/cm + organic matter	100	0.0	89	4.1	NS
High EC Control @ 13,550 uS/cm + organic matter	98	2.5	95	2.9	NS
High EC Control @ 21,150 uS/cm + organic matter	95	2.9	98	2.5	NS
Rough and Ready DWR station, Stockton	100	0.0	100	0.0	NS
Suisun Slough at Rush Ranch	98	2.5	98	2.5	NS
Sacramento River at Hood DWR Station[3]	97	2.8	100	0.0	NS
Napa River at River Park Blvd.[5]	89	6.8	82*	2.7	NS
Suisun Bay off Chipps Island (508)	100	0.0	96	2.6	NS
Montezuma Slough at Nurse Slough (609)	95	2.8	98	2.5	NS
Carquinez Strait, West of Benicia army dock (405)[5]	85	6.5	89*	4.2	NS
Grizzly Bay at Dolphin (602)[4]	90	7.1	100	0.0	NS
Field Dup.: Rough and Ready DWR station, Stockton	100	0.0	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.045	0.008	0.040***	0.003	NS
DIEPAMHR + organic matter	0.065	0.009	0.078	0.001	NS
Low EC Control @ 139.6 uS/cm + organic matter	0.082	0.013	0.085	0.005	NS
High EC Control @ 13,550 uS/cm + organic matter	0.080	0.005	0.075	0.005	NS
High EC Control @ 21,150 uS/cm + organic matter	0.062	0.007	0.052***	0.002	NS
Rough and Ready DWR station, Stockton	0.103	0.008	0.102	0.007	NS
Suisun Slough at Rush Ranch	0.088	0.007	0.099	0.006	NS
Sacramento River at Hood DWR Station[3]	0.080	0.003	0.066*	0.007	NS
Napa River at River Park Blvd.[5,6]	0.028**	0.005	0.034*	0.006	NS
Suisun Bay off Chipps Island (508)	0.082	0.003	0.088	0.007	NS
Montezuma Slough at Nurse Slough (609)	0.110	0.010	0.083	0.006	NS

Carquinez Strait, West of Benicia army dock (405)[5]	0.061	0.004	0.053	0.008	NS
Grizzly Bay at Dolphin (602)[4]	0.094	0.007	0.085	0.016	NS
Field Dup.: Rough and Ready DWR station, Stockton	0.125	0.007	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

***: $P < 0.001$

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

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

4. This high conductivity sample was compared to the High EC Control @ 13,550 uS/cm.

5. These high conductivity samples were compared to the High EC Control @ 21,150 uS/cm.

6. The Napa River sample showed significantly lower weight compared to the High EC Control @ 21,150 uS/cm, but not compared to the DIEPAMHR without organic matter.

Table B 26-2. Summary of 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 6/17/2008-6/18/2008.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Rough and Ready DWR station, Stockton	739	23.0	7.27	8.2	16.1	0.10	0.001
Suisun Slough at Rush Ranch	9250	16.7	7.23	5.1	45.0	0.14	0.001
Sacramento River at Hood DWR Station	163	21.4	7.15	7.2	12.3	0.33	0.002
Napa River at River Park Blvd.	20740	21.1	7.61	6.0	27.5	0.12	0.001
Suisun Bay off Chipps Island (508)	4542	19.8	7.60	10.3	19.1	0.06	0.001
Montezuma Slough at Nurse Slough (609)	9090	21.3	7.30	9.7	28.4	0.04	0.000
Carquinez Strait, West of Benicia army dock (405)	20420	20.1	7.50	10.2	42.9	0.07	0.001
Grizzly Bay at Dolphin (602)	12850	20.7	7.80	10.3	24.7	0.05	0.001
Field Dup.: Rough and Ready DWR station, Stockton	739	23.0	7.27	8.2	15.7	0.01	0.000

Table B 26-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 6/19/08 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 6/17/08 - 6/18/08.

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	22.7	23.8	7.4	8.0	7.65	8.18	104	60	-
DIEPAMHR + organic matter	370	22.7	24.5	7.3	8.0	7.61	8.25	104	60	-
Low EC Control @ 139.6 uS/cm + organic matter	165	22.7	24.7	7.3	8.6	7.38	7.97	44	18	-
High EC Control @ 13,550 uS/cm + organic matter	13515	22.7	24.8	7.2	8.5	7.56	7.96	1640	78	-
High EC Control @ 21,150 uS/cm + organic matter	21160	22.7	24.5	6.9	7.8	7.56	8.06	2480	90	-
Rough and Ready DWR station, Stockton	609	22.7	24.2	7.3	8.5	7.82	8.00	132	86	0.004
Suisun Slough at Rush Ranch	9080	22.6	24.1	7.2	8.5	7.79	8.15	1400	154	0.003
Sacramento River at Hood DWR Station	171	22.6	24.3	7.1	8.6	7.48	7.90	48	48	0.013
Napa River at River Park Blvd.	20455	22.8	24.5	6.4	7.9	7.80	7.90	3120	148	0.003
Suisun Bay off Chipps Island (508)	4524	22.7	24.4	7.2	8.6	7.69	8.01	528	80	0.002
Montezuma Slough at Nurse Slough (609)	8820	22.7	24.3	7.2	8.3	7.77	7.96	1160	114	0.001
Carquinez Strait, West of Benicia army dock (405)	20140	22.6	24.1	6.8	8.0	7.64	7.85	1960	98	0.002

Grizzly Bay at Dolphin (602)	12600	22.5	24.3	7.2	8.3	7.63	7.90	1640	90	0.001
Field Dup.: Rough and Ready DWR station, Stockton	628	22.6	24.4	7.1	8.4	7.81	8.10	140	84	0.001
DIEPAMHR + 25 ppb PBO	381	22.7	23.8	7.2	8.6	7.66	8.22	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	395	22.6	24.6	7.3	8.4	7.65	8.19	-	-	-
Low EC Control @ 139.6 uS/cm + organic matter + 25 ppb PBO	187	22.6	24.5	7.4	8.5	7.23	8.04	-	-	-
High EC Control @ 13,550 uS/cm + organic matter + 25 ppb PBO	13390	22.6	24.6	7.1	8.1	7.55	7.98	-	-	-
High EC Control @ 21,150 uS/cm + organic matter + 25 ppb PBO	20890	22.7	24.8	7.1	8.0	7.58	8.06	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	629	22.6	23.9	7.3	8.7	7.77	8.03	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	9225	22.6	23.9	7.1	8.2	7.80	8.12	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	187.05	22.7	24.0	7.2	8.4	7.51	7.96	-	-	-
Napa River at River Park Blvd. + 25 ppb PBO	20330	22.7	24.0	6.8	7.7	7.77	7.93	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	4462	22.7	23.9	7.3	8.6	7.69	8.02	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	9030	23.0	24.4	7.2	8.4	7.80	7.96	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	20100	22.6	24.3	6.9	8.1	7.64	7.92	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	12575	23.8	24.2	7.3	8.7	7.64	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 B 27-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 6/20/08 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 6/19/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	87	5.1	95	5.0	NS
DIEPAMHR + organic matter	86	5.4	82	4.6	NS
Low EC Control @ 209 uS+ organic matter	86	7.0	83	7.5	NS
Upper Cache Slough at mouth of Ulatis Creek	94	5.6	95	2.9	NS
Sacramento R. Deep Water Channel, Light 55	95	5.0	100	0.0	NS
Confluence of Lindsey Sl. And Cache Sl.[3]	92	2.6	98	2.5	NS
San Joaquin River at Potato Slough (815)	100	0.0	100	0.0	NS
Sacramento River at tip of Grand Island (711)[3]	95	2.9	95	2.9	NS
Old River, western arm at railroad bridge (902)	95	2.9	90	4.6	NS
Old River at mouth of Holland Cut (915)	100	0.0	100	0.0	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.073	0.004	0.060*	0.005	NS
DIEPAMHR + organic matter	0.088	0.009	0.083	0.007	NS
Low EC Control @ 209 uS + organic matter	0.089	0.012	0.089	0.025	NS
Upper Cache Slough at mouth of Ulatis Creek	0.125	0.009	0.101	0.007	NS
Sacramento R. Deep Water Channel, Light 55	0.129	0.008	0.140	0.019	NS
Confluence of Lindsey Sl. And Cache Sl.[3]	0.124	0.012	0.132	0.006	NS

San Joaquin River at Potato Slough (815)	0.131	0.011	0.126	0.009	NS
Sacramento River at tip of Grand Island (711)[3]	0.106	0.005	0.142	0.017	NS
Old River, western arm at railroad bridge (902)	0.129	0.009	0.137	0.021	NS
Old River at mouth of Holland Cut (915)	0.157	0.010	0.113	0.011	S* (72%)

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard statistical protocols.

*: $P < 0.05$

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

3. These low conductivity samples were compared to the Low EC Control.

Table B 27-2. Summary of 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 6/19/2008.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Upper Cache Slough at mouth of Ulatis Creek	243	22.0	8.00	9.2	79.9	0.10	0.004
Sacramento R. Deep Water Channel, Light 55	291	22.9	7.50	8.9	41.2	0.05	0.001
Confluence of Lindsey Sl. And Cache Sl.	214	21.4	7.80	9.3	49.6	0.10	0.003
San Joaquin River at Potato Slough (815)	244	22.6	7.60	8.8	12.0	0.10	0.002
Sacramento River at tip of Grand Island (711)	146	22.8	7.50	8.5	16.0	0.58	0.008
Old River, western arm at railroad bridge (902)	313	23.5	7.70	8.0	15.8	0.03	0.001
Old River at mouth of Holland Cut (915)	351	24.3	7.60	8.3	11.6	0.03	0.001

Table B 27-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 6/20/08 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 6/19/08.

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	22.1	24.5	7.0	8.2	7.57	8.09	104	60	-
DIEPAMHR + organic matter	344	22.1	24.3	7.3	8.2	7.51	8.08	104	60	-
Low EC Control + organic matter	240	22.0	24.8	7.0	8.6	7.52	7.98	56	36	-
Upper Cache Slough at mouth of Ulatis Creek	230	22.0	24.6	7.4	8.6	7.73	8.10	80	80	0.006
Sacramento R. Deep Water Channel, Light 55	296	22.0	24.7	7.1	8.4	7.91	8.06	92	86	0.003
Confluence of Lindsey Sl. And Cache Sl.	207	22.0	24.1	7.3	8.3	7.80	8.07	72	74	0.006
San Joaquin River at Potato Slough (815)	256	22.1	24.8	7.2	8.5	7.74	7.91	72	71	0.004
Sacramento River at tip of Grand Island (711)	164	22.0	24.7	6.9	8.1	7.65	7.76	200	53	0.016
Old River, western arm at railroad bridge (902)	346	22.1	24.7	7.5	8.5	7.81	8.08	92	74	0.001
Old River at mouth of Holland Cut (915)	396	22.1	24.7	7.3	8.2	7.81	8.13	88	166	0.002
DIEPAMHR + 25 ppb PBO	385	22.1	24.4	7.1	8.0	7.70	8.10	-	-	-
DIEPAMHR + organic matter + 25 ppb PBO	393	22.0	24.4	7.1	8.2	7.73	8.11	-	-	-
Low EC Control + organic matter + 25 ppb PBO	227	22.0	24.2	7.3	8.9	7.43	8.00	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	268	22.1	24.4	7.4	8.4	7.87	8.04	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	332	22.1	24.2	7.1	8.6	7.90	8.18	-	-	-
Confluence of Lindsey Sl. And Cache Sl. + 25 ppb PBO	250	22.0	24.3	7.3	8.3	7.84	8.04	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	239	22.0	24.3	7.1	8.1	7.70	8.63	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	134	22.0	24.2	7.1	8.2	7.61	7.90	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	329	22.0	24.1	7.6	8.3	7.70	8.02	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	334	22.1	24.2	7.2	8.3	7.82	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 B 28-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 07/03/2008 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 07/01/2008.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	94	3.2	98	2.5	NS
DIEPAMHR + POM	98	2.5	98	2.5	NS
Low EC control @ 203.1 uS/cm + POM	93	4.8	95	3.1	NS
High EC control @ 24430 uS/cm + POM	95	2.9	90	5.8	NS
Rough and Ready, DWR Station, Stockton	95	3.1	100	0.0	NS
Suisun at Rush Ranch[4]	95	4.5	95	4.5	NS
Sacramento River at Hood DWR Station[3]	95	2.9	100	0.0	NS
Napa River, near River Park Blvd.[4]	98	2.5	85	8.7	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.072	0.007	0.051	0.010	NS
DIEPAMHR + POM	0.102	0.008	0.089	0.011	NS
Low EC control @ 203.1 uS/cm + POM	0.127	0.008	0.083	0.006	S (65%)***
High EC control @ 24430 uS/cm + POM	0.068**	0.003	0.061	0.012	NS
Rough and Ready, DWR Station, Stockton	0.130	0.012	0.135	0.008	NS
Suisun at Rush Ranch[4]	0.119	0.013	0.105	0.004	NS
Sacramento River at Hood DWR Station[3]	0.103	0.018	0.101	0.007	NS
Napa River, near River Park Blvd.[4]	0.060	0.006	0.056	0.007	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

**: $P < 0.01$

***: $P < 0.001$

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

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

4. This high conductivity sample was compared to the High EC Control.

Table B 28-2. Summary of 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 07/01/2008.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Rough and Ready DWR station, Stockton	695	24.4	7.37	7.9	16.6	0.08	0.001
Suisun at Rush Ranch	12860	17.2	7.08	5.9	77.1	0.16	0.000
Sacramento River at Hood DWR Station	215	22.1	7.26	7.5	19.5	0.54	0.004
Napa River, near River Park Blvd.	22970	21.6	7.23	5.4	24.8	0.08	0.000

Table B 28-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 07/03/2008 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 07/01/2008.

Treatment	Laboratory Chemistry									
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH	Hardness (mg/L as $CaCO_3$)	Alkalinity (mg/L as $CaCO_3$)	Unionized Ammonia (mg/L)[1]
DIEPAMHR	349	23.1	24.7	7.1	8.2	7.61	8.11	100	56	-
DIEPAMHR + POM	345	23.0	24.6	6.9	8.0	7.56	8.10	100	56	-
Low EC control @ 203.1 uS/cm + POM	221	23.1	24.7	7.0	8.3	7.37	7.98	64	36	-
High EC control @ 24430 uS/cm + POM	23790	23.1	24.8	6.9	8.8	7.66	7.88	2840	90	-
Rough and Ready DWR station, Stockton	695	23.1	24.6	7.0	8.7	7.41	8.03	136	84	0.001
Suisun at Rush Ranch	10605	22.9	24.2	7.0	8.2	7.32	8.13	1640	136	0.001
Sacramento River at Hood DWR Station	188	22.9	24.8	6.6	8.4	7.33	7.82	56	62	0.006
Napa River, near River Park Blvd.	13484	23.3	24.5	6.4	8.0	7.49	7.97	2680	144	0.001
DIEPAMHR + 25 ppb PBO	601	23.4	25.3	6.6	8.2	7.60	7.99	-	-	-
DIEPAMHR + POM + 25 ppb PBO	352	22.2	23.4	6.9	8.2	7.63	7.99	-	-	-
Low EC control @ 203.1 uS/cm + POM + 25 ppb PBO	223	23.4	24.3	6.8	8.6	7.45	7.86	-	-	-
High EC control @ 24430 uS/cm + POM + 25 ppb PBO	23930	23.9	25.1	6.4	8.0	7.62	7.89	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	692	23.5	25.2	7.2	8.3	7.38	8.10	-	-	-
Suisun at Rush Ranch + 25 ppb PBO	10740	23.9	25.1	6.8	8.0	7.16	8.06	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	184	24.1	24.5	6.8	8.4	7.36	7.81	-	-	-
Napa River, near River Park Blvd. + 25 ppb PBO	13615	24.8	25.3	6.4	8.8	7.43	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 B 29-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 7/04/08 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 07/03/2008.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	98	2.5	92	4.8	NS
DIEPAMHR + POM	95	2.9	98	2.5	NS
Low EC Control @ 159.2 uS/cm + POM	86	5.8	98	2.3	NS
High EC Control @ 16.17 mS/cm + POM	100	0.0	98	2.5	NS
High EC Control @ 21.02 mS/cm + POM	98	2.5	90	4.1	NS
Sacramento River at tip of Grand Island (711)[3]	100	0.0	88	12.5	NS
Sacramento R. Deep Water Channel, Light 55	100	0.0	98	2.5	NS
Old River at mouth of Holland Cut (915)	100	0.0	100	0.0	NS

Grizzly Bay at Dolphin (602)[4]	100	0.0	100	0.0	NS
Montezuma Slough at Nurse Slough (609)	95	2.9	100	0.0	NS
Suisun Bay off Chipps Island (508)	98	2.3	100	0.0	NS
Old River, western arm at railroad bridge (902)	100	0.0	100	0.0	NS
San Joaquin River at Potato Slough (815)[3]	100	0.0	100	0.0	NS
Carquinez Strait, West of Benicia army dock (405)[5]	98	2.5	90	4.1	NS
Bottle Blank: DIEPAMHR	95	2.8	-	-	NA
Field dup: Old River, western arm at railroad bridge (902)	98	2.5	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.118	0.016	0.066	0.011	S (56%)*
DIEPAMHR + POM	0.106	0.004	0.091	0.009	NS
Low EC Control @ 159.2 uS/cm + POM	0.105	0.017	0.098	0.003	NS
High EC Control @ 16.17 mS/cm + POM	0.108	0.005	0.066	0.016	S (61%)*
High EC Control @ 21.02 mS/cm + POM	0.093*	0.003	0.059*	0.004	S (63%)***
Sacramento River at tip of Grand Island (711)[3]	0.080	0.006	0.028***	0.005	S (35%)***
Sacramento R. Deep Water Channel, Light 55	0.106	0.025	0.068	0.012	NS
Old River at mouth of Holland Cut (915)	0.107	0.022	0.090	0.005	NS
Grizzly Bay at Dolphin (602)[4]	0.103	0.005	0.048	0.014	S (46%)**
Montezuma Slough at Nurse Slough (609)	0.104	0.021	0.050*	0.012	NS
Suisun Bay off Chipps Island (508)	0.068**	0.008	0.076	0.022	NS
Old River, western arm at railroad bridge (902)	0.136	0.012	0.067	0.011	S (49%)**
San Joaquin River at Potato Slough (815)[3]	0.123	0.022	0.056**	0.011	S (46%)*
Carquinez Strait, West of Benicia army dock (405)[5]	0.062	0.019	0.053	0.008	NS
Bottle Blank: DIEPAMHR	0.052*	0.018	-	-	NA
Field dup: Old River, western arm at railroad bridge (902)	0.104	0.018	-	-	NA

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

Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

***: $P < 0.001$

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.
3. These low conductivity samples were compared to the Low EC Control.
4. These High conductivity samples were compared to the High EC Control @ 16.17 mS/cm.
5. These High conductivity samples were compared to the High EC Control @ 21.02 mS/cm.

Table B 29-2. Summary of 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 07/03/2008.

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)	-	-	-	-	13.7	0.17	-
Sacramento R. Deep Water Channel, Light 55	-	-	-	-	61.4	0.05	-
Old River at mouth of Holland Cut (915)	315	24.6	7.14	8.0	12.8	0.01	0.000

Grizzly Bay at Dolphin (602)	14760	20.4	7.76	9.3	130.7	0.13	0.002
Montezuma Slough at Nurse Slough (609)	9180	21.5	7.41	8.6	28.8	0.03	0.000
Suisun Bay off Chipps Island (508)	7580	19.9	7.49	9.3	35.0	0.04	0.000
Old River, western arm at railroad bridge (902)	293	23.3	7.10	8.2	16.4	0.04	0.000
San Joaquin River at Potato Slough (815)	218	22.7	7.41	8.6	11.4	0.06	0.001
Carquinez Strait, West of Benicia army dock (405)	20060	19.5	7.54	9.3	237.3	0.22	0.002
Bottle Blank: DIEPAMHR	-	-	-	-	0.3	0.00	-
Field dup: Old River, western arm at railroad bridge (902)	293	23.3	7.10	8.2	17.3	0.05	0.000

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	23.9	6.9	8.2	7.59	8.20	100	56	-
DIEPAMHR + POM	349	22.7	23.9	7.0	8.2	7.55	8.14	100	56	-
Low EC Control @ 159.2 uS/cm + POM	181	22.8	23.9	6.8	8.6	7.24	7.92	48	28	-
High EC Control @ 16.17 mS/cm + POM	15540	23.1	23.8	6.7	8.9	7.60	8.01	1960	84	-
High EC Control @ 21.02 mS/cm + POM	20310	22.9	23.8	6.2	8.4	7.63	8.01	3000	94	-
Sacramento River at tip of Grand Island (711)	181	23.1	24.1	6.3	8.4	7.53	7.94	64	62	0.007
Sacramento R. Deep Water Channel, Light 55	305	22.8	24.3	6.8	8.9	7.71	8.07	88	84	0.003
Old River at mouth of Holland Cut (915)	321	22.9	24.2	6.8	8.7	7.63	7.92	80	70	0.000
Grizzly Bay at Dolphin (602)	14460	23.1	24.1	6.7	8.5	7.64	7.84	2240	92	0.003
Montezuma Slough at Nurse Slough (609)	9110	23.1	24.3	6.8	8.3	7.77	7.92	1160	104	0.001
Suisun Bay off Chipps Island (508)	7650	23.1	24.3	6.8	8.4	7.55	7.91	960	86	0.001
Old River, western arm at railroad bridge (902)	295	23.0	24.1	6.9	8.5	7.68	8.03	76	66	0.002
San Joaquin River at Potato Slough (815)	224	23.1	24.2	7.0	8.8	7.63	7.95	72	66	0.003
Carquinez Strait, West of Benicia army dock (405)	18220	23.4	23.6	6.6	8.1	7.61	7.76	2720	98	0.004
Bottle Blank: DIEPAMHR	357	23.3	24.2	6.9	8.6	7.57	8.10	104	62	0.000
Field dup: Old River, western arm at railroad bridge (902)	309	23.4	24.5	6.7	8.5	7.60	7.91	72	70	0.002
DIEPAMHR + 25 ppb PBO	351	23.4	23.5	6.8	8.2	7.53	8.10	-	-	-
DIEPAMHR + POM + 25 ppb PBO	344	23.5	23.5	6.9	8.2	7.59	8.09	-	-	-
Low EC Control @ 159.2 uS/cm + POM + 25 ppb PBO	170	23.4	23.4	6.7	8.8	7.30	7.88	-	-	-
High EC Control @ 16.17 mS/cm + POM + 25 ppb PBO	15275	23.3	23.4	6.3	8.2	7.58	8.00	-	-	-
High EC Control @ 21.02 mS/cm + POM + 25 ppb PBO	20220	23.3	23.5	6.0	8.3	7.57	8.01	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	177	23.3	23.5	6.6	8.8	7.49	7.97	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	295	23.3	23.6	6.8	8.8	7.67	8.08	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	307	23.3	23.4	6.7	8.4	7.61	7.87	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	14335	23.2	23.7	6.1	8.3	7.40	7.87	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	8935	23.3	23.9	6.4	8.5	7.69	7.85	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	7545	23.2	23.7	6.5	8.9	7.49	7.92	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	310	23.4	23.7	6.9	8.7	7.61	7.93	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	224	23.3	24.2	6.7	8.9	7.57	7.91	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	18935	23.3	24.1	6.4	8.6	7.59	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 B 30-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 7/09/2008 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 7/08/2008.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	100	0.0	95	2.9	NS
DIEPAMHR + POM	100	0.0	92	2.6	S (92%)**
Low EC Control @ 179.5 uS/cm + POM	98	2.5	95	2.9	NS
Confluence of Lindsey Sl. and Cache Sl.[3]	98	2.5	90	10.0	NS
Upper Cache Slough at mouth of Ulatis Creek[3]	100	0.0	100	0.0	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.071	0.002	0.036*	0.002	S (51%)***
DIEPAMHR + POM	0.083	0.004	0.060	0.011	NS
Low EC Control @ 179.5 uS/cm + POM	0.085	0.008	0.049	0.008	S (58%)**
Confluence of Lindsey Sl. and Cache Sl.[3]	0.087	0.007	0.071	0.007	NS
Upper Cache Slough at mouth of Ulatis Creek[3]	0.089	0.009	0.078	0.006	NS

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

Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

***: $P < 0.001$

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

3. These low conductivity samples were compared to the Low EC control @ 179.5 uS/cm.

Table B 30-2. Summary of 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 7/09/2008.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (^{o}C)	pH	DO (mg/L)			
Confluence of Lindsey Sl. and Cache Sl.	167	24.3	7.00	7.5	14.3	0.15	0.001
Upper Cache Slough at mouth of Ulatis Creek	197	24.7	7.76	9.3	40.2	0.05	0.001

Table B 30-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 7/09/2008 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 7/09/2008.

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 (^{o}C)	Max Temp (^{o}C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	365	21.9	24.1	7.3	8.3	7.86	8.13	100	56	-
DIEPAMHR + POM	362	22.2	24.5	7.3	8.1	7.80	8.12	100	56	-
Low EC Control @ 179.5 uS/cm + POM	190	21.9	24.5	7.3	8.1	7.55	7.97	48	30	-
Confluence of Lindsey Sl. and Cache Sl.	173	22.8	24.2	7.2	8.4	7.77	8.14	56	62	0.010
Upper Cache Slough at mouth of Ulatis Creek	198	22.1	23.9	7.4	8.7	7.88	8.20	64	70	0.004
DIEPAMHR + 25 ppb PBO	354	23.0	24.5	7.2	8.0	7.85	8.15	-	-	-
DIEPAMHR + POM + 25 ppb PBO	358	22.8	24.5	7.2	8.2	7.81	8.14	-	-	-
Low EC Control @ 179.5 uS/cm + POM + 25 ppb PBO	188	23.4	24.5	7.4	8.4	7.52	7.96	-	-	-
Confluence of Lindsey Sl. and Cache Sl. + 25 ppb PBO	171	23.1	24.5	7.1	8.2	7.72	8.12	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	206	23.8	24.5	7.1	8.4	7.86	8.19	-	-	-

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 B 31-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 7/17/08 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 7/15/08 - 7/16/08.

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 + POM	63*	3.4	80	9.0	NS
Low EC Control + POM	44*	14.0	66	16.0	NS
High EC Control @ 13.09 mS/cm + POM	100	0.0	83	14.4	NS
High EC Control @ 17.81 mS/cm + POM	100	0.0	78	9.8	NS
High EC Control @ 27.30 mS/cm + POM	87	6.2	19*	6.6	S*** (22%)
Suisun Slough at Rush Ranch[4]	100	0.0	98	2.5	NS
Rough and Ready DWR station, Stockton	98	2.5	98	2.3	NS
Sacramento River at Hood DWR Station[3]	98	2.5	92	2.7	NS
Napa River at River Park Blvd.[6]	90	6.7	42	15.0	S* (47%)
Carquinez Strait, West of Benicia army dock (405)[6]	93	4.8	91	3.0	NS
Suisun Bay off Chipps Island (508)	100	0.0	90	4.1	NS
Grizzly Bay at Dolphin (602)[5]	95	3.1	76	9.2	NS
Montezuma Slough at Nurse Slough (609)[4]	100	0.0	85	6.5	NS
Trip Blank: DIEPAMHR	95	2.9	-	-	NA
Bottle Blank: DIEPAMHR	98	2.3	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.053	0.006	0.033	0.008	NS
DIEPAMHR + POM	0.067	0.020	0.051	0.003	NS
Low EC Control + POM	0.075	0.006	0.053	0.008	NS
High EC Control @ 13.09 mS/cm + POM	0.081	0.007	0.047	0.010	S* (58%)
High EC Control @ 17.81 mS/cm + POM	0.065	0.002	0.046	0.010	NS
High EC Control @ 27.30 mS/cm + POM	0.044	0.003	0.069	0.014	NS
Suisun Slough at Rush Ranch[4]	0.071	0.008	0.083	0.007	NS
Rough and Ready DWR station, Stockton	0.098	0.009	0.084	0.005	NS
Sacramento River at Hood DWR Station[3]	0.089	0.006	0.048	0.003	S*** (54%)
Napa River at River Park Blvd.[6]	0.034*	0.003	0.043	0.012	NS
Carquinez Strait, West of Benicia army dock (405)[6]	0.068	0.006	0.056	0.001	NS
Suisun Bay off Chipps Island (508)	0.089	0.011	0.061	0.006	NS
Grizzly Bay at Dolphin (602)[5]	0.065	0.005	0.051	0.007	NS
Montezuma Slough at Nurse Slough (609)[4]	0.093	0.007	0.047	0.008	S** (51%)
Trip Blank: DIEPAMHR	0.063	0.002	-	-	NA
Bottle Blank: DIEPAMHR	0.066	0.005	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. All comparisons were performed using USEPA standard single-concentration statistical protocols.

$P < 0.05$: *

$P < 0.01$: **

$P < 0.001$: ***

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

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

4. This high conductivity sample was compared to the High EC Control @ 13.09 mS/cm.

5. This high conductivity sample was compared to the High EC Control @ 17.81 mS/cm.

6. This high conductivity sample was compared to the High EC Control @ 27.3 mS/cm.

Table B 31-2. Summary of 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 7/15/08 - 7/16/08.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Suisun Slough at Rush Ranch	11970	18.2	7.24	5.1	153.7	0.21	0.001
Rough and Ready DWR station, Stockton	776	25.7	6.92	8.1	17.2	0.10	0.000
Sacramento River at Hood DWR Station	292	22.9	7.08	7.4	13.3	0.51	0.003
Napa River at River Park Blvd.	25790	23.4	7.19	5.8	32.4	0.07	0.000
Carquinez Strait, West of Benicia army dock (405)	21810	20.2	7.75	8.3	43.1	0.12	0.002
Suisun Bay off Chipps Island (508)	6620	20.5	7.62	8.9	15.2	0.07	0.001
Grizzly Bay at Dolphin (602)	16870	19.5	7.63	8.6	305.7	0.24	0.003
Montezuma Slough at Nurse Slough (609)	10000	21.7	7.47	8.5	15.4	0.05	0.000
Trip Blank: DIEPAMHR	365	23.2	8.3	7.9	0.6	0.03	0.003

Table B 31-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 7/17/08 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 7/15/08 - 7/16/08.

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	22.0	24.2	7.5	8.7	7.72	8.40	100	56	-
DIEPAMHR + POM	341	22.0	24.2	7.2	8.4	7.53	8.51	100	56	-
Low EC Control + POM	171	22.1	24.3	6.9	8.8	7.30	8.04	44	30	-
High EC Control @ 13.09 mS/cm + POM	12415	22.0	24.3	6.9	8.3	7.63	8.26	1440	80	-
High EC Control @ 17.81 mS/cm + POM	17120	22.0	24.2	6.9	8.2	7.59	8.33	1960	90	-
High EC Control @ 27.30 mS/cm + POM	26310	22.0	24.1	6.5	8.7	7.61	8.23	3040	98	-
Suisun Slough at Rush Ranch	11825	22.1	24.2	6.7	8.4	7.76	8.75	1480	144	0.002
Rough and Ready DWR station, Stockton	694	22.0	24.3	7.0	8.7	7.79	8.73	138	90	0.004
Sacramento River at Hood DWR Station	178	22.0	24.3	6.6	8.7	7.56	8.57	28	62	0.013
Napa River at River Park Blvd.	24500	22.1	24.2	6.5	8.0	7.70	8.66	3160	140	0.000
Carquinez Strait, West of Benicia army dock (405)	20610	22.1	24.3	6.7	8.3	7.59	8.39	2360	100	0.002
Suisun Bay off Chipps Island (508)	6320	22.0	24.2	6.9	8.6	7.69	8.60	680	72	0.003
Grizzly Bay at Dolphin (602)	15920	22.1	24.3	6.8	8.7	7.54	8.58	1800	94	0.003
Montezuma Slough at Nurse Slough (609)	9560	22.1	24.1	7.1	8.6	7.78	8.51	1640	98	0.001
Trip Blank: DIEPAMHR	376	22.1	24.1	7.7	8.7	7.80	8.55	104	62	0.001
Bottle Blank: DIEPAMHR	355	22.1	24.1	7.6	8.9	7.77	8.52	-	-	-
DIEPAMHR + 25 ppb PBO	353	22.1	24.1	7.2	8.6	7.69	8.41	-	-	-
DIEPAMHR + POM + 25 ppb PBO	350	22.1	24.1	6.9	8.7	7.53	8.62	-	-	-
Low EC Control + POM + 25 ppb PBO	186	22.1	24.1	6.9	8.9	7.27	8.22	-	-	-
High EC Control @ 13.09 mS/cm + POM + 25 ppb PBO	12540	22.2	24.4	6.8	8.7	7.63	8.36	-	-	-
High EC Control @ 17.81 mS/cm + POM + 25 ppb PBO	17085	22.2	24.4	6.8	8.4	7.60	8.30	-	-	-
High EC Control @ 27.30 mS/cm + POM + 25 ppb PBO	25100	22.1	24.3	6.8	8.3	7.66	8.54	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	11785	22.2	24.3	6.6	8.4	7.70	8.87	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	690	22.2	24.3	6.9	9.0	7.78	8.79	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	186	22.2	24.3	6.5	8.8	7.56	8.63	-	-	-
Napa River at River Park Blvd. + 25 ppb PBO	25695	22.3	24.2	6.5	8.2	7.69	8.56	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	20695	22.6	24.2	6.6	8.2	7.62	8.40	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	6455	22.5	24.2	7.0	8.8	7.64	8.44	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	16395	23.2	24.2	6.9	8.7	7.58	8.58	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	9710	22.8	24.2	7.1	8.8	7.76	8.55	-	-	-

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 B 32-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 7/18/08 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 7/17/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	98	2.5	100	0.0	NS
DIEPAMHR + POM	92	2.7	98	2.5	NS
Low EC Control @ 158.6 uS/cm + POM	64**	6.8	77	8.5	NS
Sacramento River at tip of Grand Island (711)	100	0.0	100	0.0	NS
Old River at mouth of Holland Cut (915)	100	0.0	100	0.0	NS
Upper Cache Slough at mouth of Ulatis Creek[3]	98	2.3	98	2.5	NS
San Joaquin River at Potato Slough (815)[3]	100	0.0	100	0.0	NS
Confluence of Lindsey Sl. And Cache Sl.[3]	93	2.4	95	2.9	NS
Old River, western arm at railroad bridge (902)	98	2.5	100	0.0	NS
Sacramento R. Deep Water Channel, Light 55[3]	100	0.0	98	2.5	NS
Trip Blank: DIEPAMHR	100	0.0	98	2.5	NS
Field Dup.: San Joaquin River at Potato Slough (815)[3]	100	0.0	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.031	0.009	0.065	0.013	NS
DIEPAMHR + POM	0.030	0.006	0.030	0.004	NS
Low EC Control @ 158.6 uS/cm + POM	0.035	0.009	0.034	0.006	NS
Sacramento River at tip of Grand Island (711)	0.050	0.012	0.051	0.005	NS
Old River at mouth of Holland Cut (915)	0.061	0.015	0.071	0.012	NS
Upper Cache Slough at mouth of Ulatis Creek[3]	0.046	0.002	0.041	0.010	NS
San Joaquin River at Potato Slough (815)[3]	0.085	0.011	0.036	0.007	S** (42%)
Confluence of Lindsey Sl. And Cache Sl.[3]	0.059	0.010	0.031	0.013	NS
Old River, western arm at railroad bridge (902)	0.076	0.006	0.063	0.011	NS
Sacramento R. Deep Water Channel, Light 55[3]	0.042	0.004	0.062	0.004	S* (148%)
Trip Blank: DIEPAMHR	0.024	0.006	0.062	0.004	NS
Field Dup.: San Joaquin River at Potato Slough (815)[3]	0.087	0.002	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

***: $P < 0.001$

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

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

Table B 32-2. Summary of 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 7/17/08.

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)	156	23.0	7.62	7.9	13.7	0.24	0.005
Old River at mouth of Holland Cut (915)	365	24.7	7.67	7.5	11.1	0.04	0.001
Upper Cache Slough at mouth of Ulatis Creek	199	20.8	7.44	8.4	90.4	0.09	0.001
San Joaquin River at Potato Slough (815)	241	23.3	7.62	8.1	8.3	0.05	0.001
Confluence of Lindsey Sl. And Cache Sl.	185	21.2	7.77	8.7	56.1	0.13	0.003
Old River, western arm at railroad bridge (902)	437	23.7	7.57	7.9	12.6	0.04	0.001
Sacramento R. Deep Water Channel, Light 55	281	23.1	7.38	8.2	55.3	0.00	0.000
Trip Blank: DIEPAMHR	-	-	-	-	0.2	-	-
Field Dup.: San Joaquin River at Potato Slough (815)	241	23.3	7.62	8.1	9.1	0.06	0.001

Table B 32-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 7/18/08 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 7/17/07.

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	335	22.4	23.8	7.2	8.4	7.78	8.14	100	56	-
DIEPAMHR + POM	330	22.4	23.4	7.1	8.3	7.66	8.15	100	56	-
Low EC Control @ 158.6 uS/cm + POM	155	22.4	23.8	7.1	8.5	7.40	8.02	40	30	-
Sacramento River at tip of Grand Island (711)	303	22.4	23.7	7.0	8.6	7.59	7.95	52	66	0.005
Old River at mouth of Holland Cut (915)	349	22.4	23.6	7.2	8.4	7.72	7.89	76	68	0.001
Upper Cache Slough at mouth of Ulatis Creek	191	22.4	23.5	7.2	8.5	7.72	8.03	72	74	0.002
San Joaquin River at Potato Slough (815)	232	22.4	23.6	7.1	8.6	7.74	7.97	60	66	0.002
Confluence of Lindsey Sl. And Cache Sl.	169	22.4	23.6	7.2	8.9	7.69	8.00	56	68	0.005
Old River, western arm at railroad bridge (902)	402	22.4	23.7	7.2	8.6	7.76	7.96	80	64	0.001
Sacramento R. Deep Water Channel, Light 55	334	22.4	23.6	7.4	8.4	7.76	8.11	104	66	0.000
Trip Blank: DIEPAMHR	233	22.4	23.6	7.3	8.7	7.74	8.00	64	62	0.002
Field Dup.: San Joaquin River at Potato Slough (815)	258	22.6	23.8	7.3	8.4	7.85	8.05	80	82	0.003
DIEPAMHR + 25 ppb PBO	332	22.4	23.6	7.5	8.4	7.77	8.14	-	-	-
DIEPAMHR + POM + 25 ppb PBO	330	22.4	23.5	7.0	8.4	7.56	8.14	-	-	-
Low EC Control @ 158.6 uS/cm + POM + 25 ppb PBO	156	22.4	23.7	6.9	8.4	7.29	7.99	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	148	22.4	23.6	6.8	8.7	7.60	7.94	-	-	-

Old River at mouth of Holland Cut (915) + 25 ppb PBO	344	22.3	23.7	7.1	8.6	7.72	7.92	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	190	22.5	23.7	7.2	8.8	7.82	8.04	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	232	22.5	23.8	7.0	8.8	7.69	7.96	-	-	-
Confluence of Lindsey Sl. And Cache Sl. + 25 ppb PBO	172	22.4	23.6	7.3	8.6	7.74	8.08	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	418	22.7	23.9	7.2	8.6	7.73	7.92	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	258	22.6	23.7	7.2	8.5	7.84	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 B 33-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 7/31/08 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 7/29/08 - 7/30/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMH	100	0.0	100	0.0	NS
DIEPAMH + POM	90	4.1	97	3.1	NS
High EC Control @ 13.48 mS/cm + POM	100	0.0	98	2.5	NS
High EC Control @ 18.09 mS/cm + POM	100	0.0	97	2.8	NS
High EC Control @ 23.60 mS/cm + POM	98	2.5	84	6.6	NS
Suisun Slough at Rush Ranch [3]	98	2.5	100	0.0	NS
Rough and Ready DWR Station, Stockton	100	0.0	100	0.0	NS
Sacramento River at Hood DWR Station	93	2.5	95	2.9	NS
Montezuma Slough at Nurse Slough (609)[3]	98	2.5	100	0.0	NS
Carquinez strait, West of Benicia army dock (405)[5]	83	3.6	61	20.9	NS
Suisun Bay off Chipps Island (508)	100	0.0	100	0.0	NS
Grizzly Bay at Dolphin (602)[4]	95	2.9	94	5.6	NS
Field Dup.: Grizzly Bay at Dolphin (602)[4]	100	0.0	-	-	NA
Bottle Blank: DIEPAMHR	95	2.9	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMH	0.066	0.008	0.057	0.008	NS
DIEPAMH + POM	0.078	0.016	0.063	0.013	NS
High EC Control @ 13.48 mS/cm + POM	0.089	0.012	0.068	0.007	NS
High EC Control @ 18.09 mS/cm + POM	0.069	0.006	0.056	0.003	NS
High EC Control @ 23.60 mS/cm + POM	0.041*	0.005	0.026*	0.005	NS
Suisun Slough at Rush Ranch [3]	0.086	0.006	0.071	0.007	NS
Rough and Ready DWR Station, Stockton	0.109	0.011	0.106	0.005	NS
Sacramento River at Hood DWR Station	0.099	0.011	0.083	0.009	NS

Montezuma Slough at Nurse Slough (609)[3]	0.106	0.010	0.066	0.011	S (62%)*
Carquinez strait, West of Benicia army dock (405)[5]	0.043	0.006	0.064	0.019	NS
Suisun Bay off Chipps Island (508)	0.096	0.016	0.074	0.010	NS
Grizzly Bay at Dolphin (602)[4]	0.069	0.006	0.061	0.007	NS
Field Dup.: Grizzly Bay at Dolphin (602)[4]	0.061	0.005	-	-	NA
Bottle Blank: DIEPAMHR	0.070	0.005	-	-	NA

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

Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

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

3. These high conductivity samples were compared to the High EC Control @ 13.48 mS/cm.

4. These high conductivity samples were compared to the High EC Control @ 18.09 mS/cm.

5. This high conductivity sample was compared to the High EC Control @ 23.60 mS/cm.

Table B 33-2. Summary of 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 7/29/08 - 7/30/08.

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 Hood DWR Station	178	23.3	7.86	7.3	11.5	0.43	0.014
Rough and Ready DWR Station, Stockton	718	25.4	7.56	7.6	16.0	0.06	0.001
Suisun Slough at Rush Ranch	13010	18.0	7.00	5.2	38.0	0.12	0.000
Suisun Bay off Chipps Island (508)	7190	21.5	6.80	8.3	10.1	0.03	0.000
Carquinez strait, West of Benicia army dock (405)	23070	19.8	6.43	8.1	28.6	0.08	0.000
Grizzly Bay at Dolphin (602)	18700	20.4	7.41	8.4	55.4	0.07	0.000
Field Dup.: Grizzly Bay at Dolphin (602)	18700	20.4	7.41	8.4	65.7	0.08	0.001
Montezuma Slough at Nurse Slough (609)	11460	21.5	6.51	8.2	17.3	0.02	0.000

Table B 33-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 7/31/08 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 7/29/08 - 7/30/08.

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			
DIEPAMH	336	22.7	23.7	7.4	8.1	7.82	8.11	100	60	-
DIEPAMH + POM	337	22.4	23.7	7.1	8.4	7.62	8.15	100	60	-
High EC Control @ 13.48 mS/cm + POM	12995	22.4	23.8	7.4	8.5	7.68	7.96	1520	78	-
High EC Control @ 18.09 mS/cm + POM	17430	22.4	23.7	7.1	8.5	7.73	7.99	2080	86	-
High EC Control @ 23.60 mS/cm + POM	22805	22.4	23.6	6.9	8.9	7.67	7.95	2720	94	-
Suisun Slough at Rush Ranch	12295	22.4	23.6	7.2	8.4	7.25	8.17	2440	146	0.001
Rough and Ready DWR Station, Stockton	708	22.4	23.7	7.5	8.5	7.88	8.31	144	92	0.002
Sacramento River at Hood DWR Station	179	22.4	23.7	7.1	8.4	7.76	8.12	60	66	0.026
Montezuma Slough at Nurse Slough (609)	10980	22.4	23.6	6.9	8.3	7.73	7.97	1320	108	0.000
Carquinez strait, West of Benicia army dock (405)	21975	22.4	23.6	7.0	8.2	7.73	7.83	2640	98	0.001
Suisun Bay off Chipps Island (508)	6555	22.4	23.7	7.1	8.4	7.72	7.94	760	76	0.001
Grizzly Bay at Dolphin (602)	17185	22.4	23.6	7.1	8.2	7.74	7.90	2000	92	0.002
Field Dup.: Grizzly Bay at Dolphin (602)	17420	22.4	23.6	7.1	8.1	7.74	7.93	2000	90	0.002
Bottle Blank: DIEPAMHR	392	22.4	23.6	7.5	8.5	7.86	8.16	100	60	0.000
DIEPAMH + 25 ppb PBO	360	22.4	23.0	6.6	8.2	7.78	8.14	-	-	-
DIEPAMH + POM + 25 ppb PBO	370	22.4	23.1	7.1	8.3	7.64	8.22	-	-	-
High EC Control @ 13.48 mS/cm + POM + 25 ppb PBO	12770	22.3	23.0	6.6	8.6	7.49	7.94	-	-	-
High EC Control @ 18.09 mS/cm + POM + 25 ppb PBO	17285	22.3	23.3	6.8	8.3	7.62	7.96	-	-	-
High EC Control @ 23.60 mS/cm + POM + 25 ppb PBO	22535	22.4	23.2	6.9	8.3	7.61	7.94	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	12255	22.4	23.3	6.9	8.4	7.93	8.15	-	-	-
Rough and Ready DWR Station, Stockton + 25 ppb PBO	723	22.2	23.1	7.4	8.5	7.83	8.29	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	197	22.2	23.4	6.7	8.5	7.82	8.09	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	10845	22.2	23.1	6.9	8.5	7.74	8.01	-	-	-
Carquinez strait, West of Benicia army dock (405) + 25 ppb PBO	21725	22.2	23.1	6.5	8.3	7.64	7.88	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	6500	22.3	23.0	6.8	8.4	7.66	7.92	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	16900	22.3	23.3	7.2	8.5	7.73	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 B 34-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 8/01/08 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 7/31/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	95	2.8	100	0.0	NS
DIEPAMHR + POM	90	7.1	97	2.8	NS
Low EC Control + POM	92	5.3	90	4.1	NS
Confluence of Lindsey Sl. And Cache Sl.[3]	95	3.1	100	0.0	NS
Sacramento River at tip of Grand Island (711)[3]	98	2.5	100	0.0	NS
San Joaquin River at Potato Slough (815)[3]	100	0.0	98	2.5	NS
Upper Cache Slough at mouth of Ulatis Creek[3]	100	0.0	100	0.0	NS
Sacramento R. Deep Water Channel, Light 55[3]	98	2.5	95	2.9	NS
Old River at mouth of Holland Cut (915)	100	0.0	98	2.5	NS
Old River, western arm at railroad bridge (902)	100	0.0	100	0.0	NS
Trip Blank: DIEPAMHR	100	0.0	-	-	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.038**	0.005	0.045	0.006	NS
DIEPAMHR + POM	0.072	0.006	0.062	0.012	NS
Low EC Control + POM	0.066	0.005	0.058	0.006	NS
Confluence of Lindsey Sl. And Cache Sl.[3]	0.069	0.008	0.052	0.003	NS
Sacramento River at tip of Grand Island (711)[3]	0.061	0.005	0.064	0.003	NS
San Joaquin River at Potato Slough (815)[3]	0.088	0.009	0.071	0.004	NS
Upper Cache Slough at mouth of Ulatis Creek[3]	0.079	0.011	0.067	0.004	NS
Sacramento R. Deep Water Channel, Light 55[3]	0.082	0.012	0.085	0.015	NS
Old River at mouth of Holland Cut (915)	0.089	0.009	0.058	0.007	S (65%)*
Old River, western arm at railroad bridge (902)	0.093	0.011	0.066	0.008	NS
Trip Blank: DIEPAMHR	0.047*	0.007	-	-	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

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

3. These low conductivity samples were compared to the Low EC Control.

Table B 34-2. Summary of 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 07/31/2008.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Confluence of Lindsey Sl. And Cache Sl.	227	20.9	6.80	9.1	55.3	0.14	0.000
Sacramento River at tip of Grand Island (711)	242	23.1	6.82	8.1	12.4	0.47	0.001
San Joaquin River at Potato Slough (815)	331	23.6	6.81	8.0	7.3	0.05	0.000
Upper Cache Slough at mouth of Ulatis Creek	200	20.3	6.76	8.3	78.9	0.08	0.000
Sacramento R. Deep Water Channel, Light 55	262	22.1	7.28	9.4	49.1	0.07	0.001
Old River at mouth of Holland Cut (915)	433	24.5	6.78	7.6	9.4	0.03	0.000
Old River, western arm at railroad bridge (902)	484	24.8	6.83	7.8	10.0	0.04	0.000
Trip Blank: DIEPAMHR	364	22.9	8.16	8.9	-	-	-

Table B 34-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 08/01/2008 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 07/31/2008.

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	332	23.6	23.6	7.5	8.8	7.76	8.04	100	60	-
DIEPAMHR + POM	332	23.5	23.5	7.5	8.6	7.76	8.05	100	60	-
Low EC Control + POM	164	23.5	23.5	7.3	8.8	7.41	7.90	52	30	-
Confluence of Lindsey Sl. And Cache Sl.	173	23.6	23.6	7.5	8.8	7.83	8.00	68	76	0.005
Sacramento River at tip of Grand Island (711)	165	23.6	23.6	7.2	8.9	7.57	8.01	60	72	0.008
San Joaquin River at Potato Slough (815)	245	23.6	23.6	7.2	8.6	7.72	8.03	72	64	0.001
Upper Cache Slough at mouth of Ulatis Creek	196	23.7	23.7	7.2	8.9	7.78	7.99	72	78	0.002
Sacramento R. Deep Water Channel, Light 55	251	23.6	23.6	7.2	8.7	7.84	8.04	80	80	0.003
Old River at mouth of Holland Cut (915)	417	23.7	23.7	7.2	8.8	7.63	7.87	84	66	0.001
Old River, western arm at railroad bridge (902)	471	23.5	23.5	7.4	8.8	7.52	7.91	84	66	0.001
Trip Blank: DIEPAMHR	334	23.3	23.3	7.6	8.8	7.76	8.05	104	56	0.000
DIEPAMHR + 25 ppb PBO	335	23.3	23.3	8.5	8.5	8.07	8.07	-	-	-
DIEPAMHR + POM + 25 ppb PBO	334	23.1	23.1	8.8	8.8	8.05	8.05	-	-	-
Low EC Control + POM + 25 ppb PBO	165	23.0	23.0	8.9	8.9	7.83	7.83	-	-	-
Confluence of Lindsey Sl. And Cache Sl. + 25 ppb PBO	173	23.0	23.0	8.9	8.9	7.85	7.85	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	167	22.9	22.9	8.6	8.6	7.59	7.59	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb	248	22.9	22.9	8.6	8.6	7.72	7.72	-	-	-

PBO										
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	191	22.9	22.9	8.8	8.8	7.81	7.81	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	246	22.8	22.8	8.8	8.8	7.93	7.93	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	410	22.8	22.8	8.5	8.5	7.66	7.66	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	463	22.7	22.7	8.4	8.4	7.65	7.65	-	-	-

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 B 35-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 8/14/08 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 8/12/08 - 8/13/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	98	2.5	98	2.5	NS
DIEPAMHR + POM	90	4.1	83	12.0	NS
Low EC Control @ 179.9 uS/cm + POM	80	0.0	70*†	12.6	NA
High EC Control @ 15.09 mS/cm + POM	98	2.5	95	2.9	NS
High EC Control @ 20.27 mS/cm + POM	100	0.0	50*†	20.8	S (50%)*
Suisun Slough at Rush Ranch[4]	16*	6.5	0*	0.0	NS
Rough and Ready DWR station, Stockton	85	15.0	100	0.0	NS
Sacramento River at Hood DWR Station[3]	98	2.5	98	2.5	NS
Montezuma Slough at Nurse Slough (609)[4]	93	6.8	100	0.0	NS
Suisun Bay off Chipps Island (508)	100	0.0	100	0.0	NS
Grizzly Bay at Dolphin (602)[5]	95	5.0	55*†	18.2	NS
Field Dup.: Suisun Bay off Chipps Island (508)	100	0.0	-	-	NA
Bottle Blank: DIEPAMHR	97	3.3	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.049	0.006	0.054	0.006	NS
DIEPAMHR + POM	0.068	0.005	0.056	0.000	NS
Low EC Control @ 179.9 uS/cm + POM	0.069	0.009	0.064	0.009	NA
High EC Control @ 15.09 mS/cm + POM	0.096	0.004	0.070	0.004	S (73%)**
High EC Control @ 20.27 mS/cm + POM	0.077	0.004	0.070	0.006	NS
Suisun Slough at Rush Ranch[4]	0.075	0.015	-	-	NA
Rough and Ready DWR station, Stockton	0.089	0.017	0.077	0.007	NS
Sacramento River at Hood DWR Station[3]	0.083	0.010	0.080	0.009	NS
Montezuma Slough at Nurse Slough (609)[4]	0.068	0.005	0.072	0.005	NS
Suisun Bay off Chipps Island (508)	0.087	0.011	0.079	0.012	NS
Grizzly Bay at Dolphin (602)[5]	0.058*	0.006	0.049*	0.006	NS
Field Dup.: Suisun Bay off Chipps Island (508)	0.083	0.003	-	-	NA
Bottle Blank: DIEPAMHR	0.072	0.003	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

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

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

4. This high conductivity sample was compare to the High EC Control @ 15.09 mS/cm.

5. This high conductivity sample was compare to the High EC Control @ 20.27 mS/cm.

†. These PBO-added samples showed lower survival compared to the DIEPAMHR + PBO control, but not compared to the PBO control with added POM.

Table B 35-2. Summary of 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 8/12/08 - 8/13/08.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Suisun Slough at Rush Ranch	14140	17.7	7.24	5.6	176.7	0.21	0.001
Rough and Ready DWR station, Stockton	697	25.4	7.64	5.8	13.1	0.07	0.002
Sacramento River at Hood DWR Station	180	23.7	7.54	7.1	9.0	0.39	0.006
Montezuma Slough at Nurse Slough (609)	12610	23.2	7.76	7.9	12.5	0.01	0.000
Suisun Bay off Chipps Island (508)	7220	21.5	7.89	8.6	6.8	0.02	0.001
Grizzly Bay at Dolphin (602)	19400	21.3	7.95	8.4	11.4	0.01	0.000
Field Dup.: Suisun Bay off Chipps Island (508)	7220	21.5	7.89	8.6	7.3	0.01	0.000

Table B 35-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 8/14/08 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 8/12/08 - 8/13/08.

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	23.3	24.5	6.9	8.0	7.66	8.08	100	58	-
DIEPAMHR + POM	344	23.3	24.4	7.0	8.2	7.42	8.07	100	58	-
Low EC Control @ 179.9 uS/cm + POM	196	23.3	24.5	6.7	8.8	7.25	7.97	48	28	-
High EC Control @ 15.09 mS/cm + POM	14690	23.4	24.4	7.0	8.6	7.51	8.00	1720	72	-
High EC Control @ 20.27 mS/cm + POM	19710	23.3	24.4	7.0	8.6	7.60	8.00	2080	80	-
Suisun Slough at Rush Ranch	13740	23.9	24.4	6.8	8.5	7.44	8.06	1680	152	0.002
Rough and Ready DWR station, Stockton	719	23.2	24.5	6.7	8.6	7.73	8.02	140	94	0.002
Sacramento River at Hood DWR Station	203	23.3	24.5	6.7	8.6	7.63	7.76	60	68	0.011
Montezuma Slough at Nurse Slough (609)	12030	23.2	24.5	7.0	8.5	7.75	7.91	1360	98	0.000
Suisun Bay off Chipps Island (508)	7180	23.3	24.5	7.0	8.6	7.69	7.91	800	70	0.001
Grizzly Bay at Dolphin (602)	18715	23.4	24.5	6.8	8.6	7.65	7.82	2120	90	0.000
Field Dup.: Suisun Bay off Chipps Island (508)	7125	23.1	24.5	7.0	8.6	7.59	7.87	800	68	0.000

Bottle Blank: DIEPAMHR	367	23.2	24.5	7.0	8.4	7.77	8.36	104	54	0.000
DIEPAMHR + 25 ppb PBO	359	23.2	24.3	7.2	8.3	7.79	8.31	-	-	-
DIEPAMHR + POM + 25 ppb PBO	349	23.1	24.3	7.2	8.3	7.54	8.15	-	-	-
Low EC Control @ 179.9 uS/cm + POM + 25 ppb PBO	200	23.2	24.3	7.0	8.7	7.28	8.14	-	-	-
High EC Control @ 15.09 mS/cm + POM + 25 ppb PBO	14430	23.1	24.4	7.1	8.7	7.59	8.02	-	-	-
High EC Control @ 20.27 mS/cm + POM + 25 ppb PBO	19780	23.1	24.4	6.9	8.3	7.68	8.02	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	13850	24.4	24.4	7.2	7.2	7.47	7.47	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	702	23.3	24.4	7.0	8.5	7.88	8.01	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	187	23.2	24.4	6.8	8.5	7.65	8.02	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	12030	23.2	24.4	7.0	8.4	7.68	7.92	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	7255	23.3	24.4	6.9	8.9	7.65	7.84	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	18810	23.3	24.4	7.0	8.8	7.68	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 B 36-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 8/15/08 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 8/14/08.

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 + POM	84*	5.8	87	7.1	NS
Low EC Control @ 177 uS/cm + POM	88	9.5	90	7.1	NS
Confluence of Lindsey Sl. and Cache Sl.[3]	95	2.9	98	2.5	NS
Sacramento River at tip of Grand Island (711)[3]	100	0.0	88	6.3	NS
San Joaquin River at Potato Slough (815)	98	2.3	100	0.0	NS
Upper Cache Slough at mouth of Ulatis Creek[3]	92	2.7	100	0.0	NS
Sacramento R. Deep Water Channel, Light 55	85*	8.7	97	3.3	NS
Old River at mouth of Holland Cut (915)[4]	98	2.5	100	0.0	NA
Old River, western arm at railroad bridge (902)	100	0.0	100	0.0	NS
Trip Blank: DIEPAMHR[5]	85	15.0	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.065	0.006	0.083	0.021	NS
DIEPAMHR + POM	0.078	0.005	0.074	0.006	NS
Low EC Control @ 177 uS/cm + POM	0.085	0.009	0.056*	0.004	S* (66%)
Confluence of Lindsey Sl. and Cache Sl.[3]	0.079	0.007	0.075	0.005	NS
Sacramento River at tip of Grand Island (711)[3]	0.078	0.007	0.062	0.004	NS

San Joaquin River at Potato Slough (815)	0.090	0.012	0.074	0.008	NS
Upper Cache Slough at mouth of Ulatis Creek[3]	0.078	0.011	0.075	0.003	NS
Sacramento R. Deep Water Channel, Light 55	0.085	0.011	0.074	0.012	NS
Old River at mouth of Holland Cut (915)[4]	0.099	0.005	0.072	0.006	NA
Old River, western arm at railroad bridge (902)	0.088	0.001	0.075	0.011	NS
Trip Blank: DIEPAMHR[5]	0.115	0.050	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

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

3. These low conductivity samples were compared to the Low EC Control.

4. Two PBO-treated replicates of sample 915 were excluded due to early mortality and high inter-replicate variability of test animal performance indicative of contamination. The PBO treatment was not included in statistical analysis.

5. Two replicates of the trip blank were excluded due to early mortality and high inter-replicate variability of test animal performance indicative of contamination. This sample was not included in statistical analysis.

Table B 36-2. Summary of 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 8/14/08.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Confluence of Lindsey Sl. and Cache Sl.	185	22.6	7.96	8.3	31.9	0.15	0.006
Sacramento River at tip of Grand Island (711)	172	24.1	7.67	7.8	7.2	0.37	0.008
San Joaquin River at Potato Slough (815)	318	24.2	7.91	7.9	4.9	0.09	0.003
Upper Cache Slough at mouth of Ulatis Creek	198	21.9	8.12	8.3	48.8	0.07	0.004
Sacramento R. Deep Water Channel, Light 55	249	22.7	8.09	8.2	37.0	0.09	0.005
Old River at mouth of Holland Cut (915)	775	25.3	7.80	7.6	7.5	0.05	0.002
Old River, western arm at railroad bridge (902)	633	24.9	7.91	7.8	6.9	0.05	0.002
Trip Blank: DIEPAMHR	350	22.1	8.03	8.2	0.2	0.02	0.001

Table B 36-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 8/15/08 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 8/14/08.

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	343	22.5	24.9	7.4	8.0	7.48	8.15	100	58	-
DIEPAMHR + POM	343	22.5	24.9	6.4	8.2	7.38	8.14	100	58	-
Low EC Control @ 177 uS/cm + POM	179	22.6	25.0	6.4	8.6	7.19	8.00	52	28	-
Confluence of Linsey Sl. And Cache Sl.	188	23.0	24.9	6.8	8.5	7.50	8.05	64	72	0.008

Sacramento River at tip of Grand Island (711)	173	22.6	24.9	5.7	8.6	7.32	8.04	60	68	0.020
San Joaquin River at Potato Slough (815)	314	23.2	24.9	7.0	8.5	7.52	8.04	76	68	0.005
Upper Cache Slough at mouth of Ulatis Creek	201	22.7	24.9	6.8	8.3	6.66	8.15	72	76	0.005
Sacramento R. Deep Water Channel, Light 55	243	23.3	24.9	6.9	8.3	7.60	8.17	80	80	0.006
Old River at mouth of Holland Cut (915)	546	22.8	25.0	6.5	8.4	7.51	7.98	96	68	0.002
Old River, western arm at railroad bridge (902)	619	23.2	24.9	7.0	8.4	7.54	8.04	104	66	0.003
Trip Blank: DIEPAMHR	344	24.9	24.9	6.8	8.3	7.48	8.13	104	58	0.001
DIEPAMHR + 25 ppb PBO	343	22.7	24.7	6.9	8.4	7.56	8.12	-	-	-
DIEPAMHR + POM + 25 ppb PBO	378	23.4	24.9	6.7	8.4	7.36	7.97	-	-	-
Low EC Control @ 177 uS/cm + POM + 25 ppb PBO	181	22.7	24.9	6.5	8.6	7.17	8.11	-	-	-
Confluence of Lindsey Sl. and Cache Sl. + 25 ppb PBO	189	23.6	25.0	6.8	8.7	7.51	7.98	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	173	22.8	25.0	5.9	8.8	7.36	8.05	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	317	23.7	25.0	6.9	8.5	7.54	8.16	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	201	23.8	25.0	6.8	8.4	7.61	8.15	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	243	22.9	25.0	7.0	8.4	7.60	7.96	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	543	23.0	25.0	6.8	8.7	7.54	8.04	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	626	23.7	24.9	6.9	8.4	7.54	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 B 37-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 8/16/2008 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 8/15/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	88	6.3	75	25.0	NS
DIEPAMHR + 1% nutrient add back	98	2.5	73	24.3	NS
High EC Control @ 13.5 uS + 1% nutrient add back	100	0.0	100	0.0	NS
Suisun Slough at Rush Ranch[3]	81	16.0	98	2.5	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.048	0.008	0.059	0.002	NS
DIEPAMHR + 1% nutrient add back	0.065	0.007	0.067	0.010	NS
High EC Control @ 13.5 uS + 1% nutrient add back	0.074	0.003	0.074	0.003	NS
Suisun Slough at Rush Ranch[3]	0.052	0.015	0.069	0.010	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.
2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.
3. This high conductivity sample was compared to the High EC Control.

Table B 37-2. Summary of 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 8/15/2008.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Suisun Slough at Rush Ranch	13350	28.0	7.72	8.0	20.7	0.04	0.001

Table B 37-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 8/16/2008 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 8/15/2008.

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	345	22.8	24.1	7.5	8.1	7.62	8.07	100	58	-
DIEPAMHR + 1% nutrient add back	344	22.9	24.3	7.2	8.2	7.55	8.02	100	58	-
High EC Control @ 13.5 uS + 1% nutrient add back	13000	22.9	23.9	7.2	8.5	7.55	7.92	1560	80	-
Suisun Slough at Rush Ranch	13015	22.9	24.2	7.2	8.9	7.56	8.07	1680	172	0.001
DIEPAMHR + 25 ppb PBO	343.9	23	23.5	7.6	8.1	7.64	8.08	-	-	-
DIEPAMHR + 25 ppb PBO + 1% nutrient add back	345.9	22.8	23.4	7.2	8.1	7.54	8.01	-	-	-
High EC Control @ 13.5 uS + 25 ppb PBO + 1% nutrient add back	12940	22.8	23.7	7.4	8.8	7.56	7.91	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	12916	22.6	23.8	7.5	8.6	7.57	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 B 38-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 8/20/2008 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 8/12/2008 - 8/14/2008.

Treatment	Survival (%)[1]	
	Unmanipulated	25 ppb PBO added

	mean	se	mean	se	vs Non-PBO[2]
DIEPAMH	92	2.8	97	2.8	NS
DIEPAMH + POM	90	4.1	95	2.8	NS
Low EC Control @ 179.9 uS + POM	85	6.5	100	0.0	NS
High EC Control @ 13.47 mS + POM	100	0.0	98	2.5	NS
High EC Control @ 20.27 mS + POM	92	4.8	98	2.5	NS
Rough and Ready DWR Station, Stockton	98	2.5	100	0.0	NS
Sacramento River at Hood DWR Station[3]	74	2.6	60*	9.1	NS
Montezuma Slough at Nurse Slough (609)[4]	100	0.0	95	2.9	NS
Suisun Bay off Chipps Island (508)	95	3.1	93	2.5	NS
Grizzly Bay at Dolphin (602)[5]	100	0.0	97	2.8	NS
Old River at mouth of Holland Cut (915)	100	0.0	80	16.8	NS
Sacramento River Deep Water Channel, Light 55	84	3.2	95	2.9	NS
Field Dup: Suisun Bay off Chipps Island (508)	100	0.0	-	-	NA
Contaminated glassware + DIEPAMH	77*	4.7	22*	9.3	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMH	0.026**	0.005	0.046	0.008	NS
DIEPAMH + POM	0.072	0.009	0.057	0.006	NS
Low EC Control @ 179.9 uS + POM	0.082	0.014	0.061	0.016	NS
High EC Control @ 13.47 mS + POM	0.074	0.004	0.052	0.011	NS
High EC Control @ 20.27 mS + POM	0.062	0.002	0.035*	0.005	S* (56%)
Rough and Ready DWR Station, Stockton	0.086	0.012	0.072	0.013	NS
Sacramento River at Hood DWR Station[3]	0.057	0.003	0.109	0.016	S* (191%)
Montezuma Slough at Nurse Slough (609)[4]	0.041**	0.005	0.054	0.001	NS
Suisun Bay off Chipps Island (508)	0.049	0.003	0.031*	0.006	S* (63%)
Grizzly Bay at Dolphin (602)[5]	0.042*	0.008	0.015	0.002	S* (36%)
Old River at mouth of Holland Cut (915)	0.091	0.007	0.076	0.006	NS
Sacramento River Deep Water Channel, Light 55	0.067	0.002	0.046	0.002	S*** (69%)
Field Dup: Suisun Bay off Chipps Island (508)	0.060†	0.003	-	-	NA
Contaminated glassware + DIEPAMH	0.071	0.003	0.032	0.010	NS

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

Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.1$

**: $P < 0.05$

***: $P < 0.01$

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

3. This low EC sample was compared to the low EC Control at 179.9 uS/cm.

4. This high EC sample was compared to the high EC Control at 13.47 mS/cm.

5. This high EC sample was compared to the high EC Control at 20.27 mS/cm.

†. The field duplicate showed heavier weights compared to the original sample collected at site 508.

Table B 38-2. Summary of 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 8/12/2008 - 8/14/2008.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Rough and Ready DWR Station, Stockton	697	25.4	7.64	5.8	13.1	0.07	0.002
Sacramento River at Hood DWR Station	180	23.7	7.54	7.1	9.0	0.39	0.006
Montezuma Slough at Nurse Slough (609)	12610	23.2	7.76	7.9	12.5	0.01	0.000
Suisun Bay off Chipps Island (508)	7220	21.5	7.89	8.6	6.8	0.02	0.001
Grizzly Bay at Dolphin (602)	19400	21.3	7.95	8.4	11.4	0.01	0.000
Old River at mouth of Holland Cut (915)	775	25.3	7.8	7.6	7.5	0.05	0.002
Sacramento River Deep Water Channel, Light 55	249	22.7	8.09	8.2	37.0	0.09	0.005
Field Dup: Suisun Bay off Chipps Island (508)	7220	21.5	7.89	8.6	7.3	0.01	0.000

Table B 38-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 8/20/2008 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 8/12/2008 - 8/14/2008.

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			
DIEPAMH	348	22.8	24.6	7.1	8.1	7.67	8.09	100	58	-
DIEPAMH + POM	343	22.8	24.6	7.0	8.3	7.65	8.09	100	58	-
Low EC Control @ 179.9 uS/cm + POM	189	22.7	24.6	7.3	8.5	7.41	7.87	48	31	-
High EC Control @ 13.47 mS/cm + POM	13090	23.0	24.6	6.9	8.3	7.48	8.00	1320	77	-
High EC Control @ 20.27 mS/cm + POM	20090	23.2	24.6	6.8	8.2	7.48	7.97	1960	85	-
Rough and Ready DWR Station, Stockton	696	23.0	24.5	7.0	8.6	7.84	8.20	140	94	0.002
Sacramento River at Hood DWR Station	214	23.8	24.5	7.2	8.6	7.58	7.88	60	68	0.014
Montezuma Slough at Nurse Slough (609)	12410	23.9	24.6	7.1	8.7	7.63	8.00	1360	98	0.000
Suisun Bay off Chipps Island (508)	7500	23.5	24.8	7.1	8.7	7.67	7.93	800	70	0.001
Grizzly Bay at Dolphin (602)	18930	23.8	24.5	7.2	8.5	7.59	7.84	2120	90	0.000
Old River at mouth of Holland Cut (915)	624	23.5	24.6	7.4	8.5	7.72	8.01	96	68	0.002
Sacramento River Deep Water Channel, Light 55	284	23.6	24.5	7.2	8.6	7.73	8.08	80	80	0.005
Field Dup: Suisun Bay off Chipps Island (508)	7140	23.8	24.6	7.2	8.6	7.65	7.94	800	68	0.000
DIEPAMH + 25 ppb PBO	376	24.2	24.6	7.2	8.4	7.73	8.09	-	-	-
DIEPAMH + POM + 25 ppb PBO	355	24.0	24.2	7.2	8.3	7.66	8.07	-	-	-
Low EC Control @ 179.9 uS/cm + POM + 25 ppb PBO	218	24.2	24.3	7.0	8.8	7.45	7.84	-	-	-
High EC Control @ 13.47 mS/cm + POM + 25 ppb PBO	13370	23.6	24.3	7.5	8.4	7.63	7.97	-	-	-
High EC Control @ 20.27 mS/cm + POM + 25 ppb PBO	20240	24.2	24.4	7.4	8.2	7.58	8.27	-	-	-
Rough and Ready DWR Station, Stockton + 25 ppb PBO	697	24.3	24.4	7.3	8.7	7.94	8.23	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	234	24.4	24.4	6.8	8.7	7.64	7.97	-	-	-
Montezuma Slough at Nurse Slough + 25 ppb PBO	12420	24.0	24.5	7.2	8.3	7.70	7.98	-	-	-
Suisun Bay off Chipps Island + 25 ppb PBO	7575	24.4	24.5	7.0	8.6	7.59	7.90	-	-	-
Grizzly Bay at Dolphin + 25 ppb PBO	19130	24.0	24.4	7.0	8.2	7.50	7.85	-	-	-
Old River at mouth of Holland Cut + 25 ppb PBO	1330	22.3	24.4	7.2	8.6	7.69	8.06	-	-	-
Sacramento River Deep Water Channel, Light 55 + 25 ppb PBO	273	24.5	24.5	7.2	8.7	7.70	8.13	-	-	-

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 B 39-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 8/28/08 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 8/26/08 - 8/27/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	80	13.5	76*	8.6	NS
DIEPAMHR + POM	80	3.9	88	5.7	NS
Low EC Control @194.4 uS/cm + POM	95	2.9	74*	3.4	S** (78%)
High EC Control @ 14.79 mS/cm + POM	100	0.0	98	2.5	NS
High EC Control @ 19.04 mS/cm + POM	100	0.0	94	5.6	NS
Sacramento River at Hood DWR Station[3,6]	72*	8.2	72	3.4	NS
Rough and Ready DWR station, Stockton	98	2.5	95	5.0	NS
Suisun Slough at Rush Ranch[4]	100	0.0	95	2.9	NS
Suisun Bay off Chipps Island (508)	100	0.0	100	0.0	NS
Montezuma Slough at Nurse Slough (609)[4]	98	2.3	98	2.5	NS
Grizzly Bay at Dolphin (602)[5]	90	7.1	100	0.0	NS
Field Dup.: Montezuma Slough at Nurse Slough (609)[4]	98	2.5	-	-	NA
Trip Blank	97	2.8	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.050	0.008	0.044	0.002	NS
DIEPAMHR + POM	0.052	0.009	0.070	0.010	NS
Low EC Control @194.4 uS/cm + POM	0.065	0.009	0.069	0.009	NS
High EC Control @ 14.79 mS/cm + POM	0.073	0.004	0.068	0.005	NS
High EC Control @ 19.04 mS/cm + POM	0.058	0.005	0.070	0.007	NS
Sacramento River at Hood DWR Station[3]	0.069	0.014	0.090	0.015	NS
Rough and Ready DWR station, Stockton	0.107	0.006	0.096	0.011	NS
Suisun Slough at Rush Ranch[4]	0.067	0.009	0.070	0.013	NS
Suisun Bay off Chipps Island (508)	0.078	0.009	0.092	0.003	NS
Montezuma Slough at Nurse Slough (609)[4]	0.082	0.009	0.084	0.003	NS
Grizzly Bay at Dolphin (602)[5]	0.066	0.005	0.078	0.003	NS
Field Dup.: Montezuma Slough at Nurse Slough (609)[4]	0.072	0.005	-	-	NA
Trip Blank	0.045	0.006	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols. Both DIEPAMHR and DIEPAMHR + POM method control treatments showed 79.7% survival, below the test acceptability criterion of 80% survival.

*: $P < 0.05$

**: $P < 0.01$

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

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

4. This high conductivity sample was compared to the High EC Control @14.79 mS/cm.

5. This high conductivity sample was compared to the High EC Control @ 19.04 mS/cm.

6. The untreated Hood sample showed significantly lower survival compared to the Low EC Control, but not compared to the Normal EC Controls with or without added POM.

Table B 39-2. Summary of 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 8/26/08 - 8/27/08.

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 Hood DWR Station	282	27.7	7.45	7.1	19.4	0.24	0.004
Rough and Ready DWR station, Stockton	845	29.8	7.58	6.3	13.4	0.06	0.002
Suisun Slough at Rush Ranch	14330	23.1	7.53	6.7	20.6	0.07	0.001
Suisun Bay off Chipps Island (508)	6490	22.2	7.80	8.2	7.3	0.01	0.000
Montezuma Slough at Nurse Slough (609)	13960	23.0	7.50	7.4	23.8	0.02	0.000
Grizzly Bay at Dolphin (602)	18280	21.8	7.71	8.0	16.7	0.05	0.001
Field Dup.: Montezuma Slough at Nurse Slough (609)	13960	23.0	7.50	7.4	18.1	0.04	0.000
Trip Blank	350	21.9	8.00	8.0	0.2	0.00	0.000

Table B 39-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 8/28/08 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 8/26/08 - 8/27/08.

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			
DIEPAMH	344	22.6	24.4	7.2	8.3	7.61	8.14	104	60	-
DIEPAMH + POM	341	22.5	24.5	7.0	8.3	7.61	8.17	104	60	-
Low EC @194.4 uS/cm + POM	205	22.5	24.5	7.2	11.2	7.43	8.02	64	36	-
High EC @ 14.79 mS/cm + POM	14135	22.4	24.5	7.0	8.6	7.54	7.94	2400	83	-
High EC @ 19.04 mS/cm + POM	18390	22.5	24.5	5.2	8.0	7.63	8.01	1560	90	-
Sacramento River at Hood DWR Station	272	22.5	24.4	7.0	8.6	7.77	8.31	72	80	0.022
Rough and Ready DWR station, Stockton	746	22.6	24.5	7.2	8.5	7.94	8.01	160	98	0.003
Suisun Slough at Rush Ranch	13865	22.4	24.5	6.8	8.6	7.73	8.06	1680	168	0.001
Suisun Bay off Chipps Island (508)	6280	22.7	24.5	6.8	8.3	7.68	7.83	600	82	0.000
Montezuma Slough at Nurse Slough (609)	13485	22.5	24.4	6.9	8.0	7.67	7.90	1720	111	0.000
Grizzly Bay at Dolphin (602)	17605	22.5	24.5	7.0	8.6	7.63	7.77	1920	95	0.001
Field Dup.: Montezuma Slough at Nurse Slough (609)	13665	22.5	24.5	7.2	8.2	7.69	7.89	1560	111	0.001
Trip Blank	420	22.5	24.5	7.2	8.6	7.70	8.18	104	62	0.000
DIEPAMH + 25 ppb PBO	348	22.4	24.5	7.0	8.4	7.66	8.22	-	-	-
DIEPAMH + POM + 25 ppb PBO	348	22.7	24.5	6.8	8.5	7.64	8.19	-	-	-
Low EC @194.4 uS/cm + POM + 25 ppb PBO	207	22.5	24.5	7.3	8.6	7.52	8.00	-	-	-
High EC @ 14.79 mS/cm + POM + 25 ppb PBO	14300	22.5	24.5	7.2	8.3	7.63	7.88	-	-	-
High EC @ 19.04 mS/cm + POM + 25 ppb PBO	18430	22.5	24.5	6.9	8.5	7.55	7.75	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	295	22.5	24.4	6.9	8.6	7.72	8.06	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	741	23.0	24.4	7.2	8.7	7.92	8.16	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	13845	22.7	24.5	7.0	8.3	7.69	8.08	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	6385	22.8	24.5	6.9	8.6	7.64	7.85	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	13570	22.9	24.5	6.9	8.1	7.65	7.88	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	17665	23.2	24.5	7.1	8.4	7.60	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 B 40-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 8/29/08 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 8/28/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	84	3.1	94	6.3	NS
DIEPAMHR + POM	80	12.2	72	8.2	NS
Low EC Control @ 203.1 uS/cm + POM	91	6.0	81	10.8	NS
Confluence of Lindsey Sl. and Cache Sl.	83	4.0	74	9.4	NS
Sacramento River at tip of Grand Island (711)	81	3.9	75	10.2	NS
San Joaquin River at Potato Slough (815)	88	5.1	88	8.8	NS
Upper Cache Slough at mouth of Ulatis Creek	84	9.4	84	3.2	NS
Sacramento R. Deep Water Channel, Light 55	93	3.9	91	6.0	NS
Old River at mouth of Holland Cut (915)[3]	94	3.6	60*	14.1	S* (64%)
Old River, western arm at railroad bridge (902)[3]	97	3.1	28	20.7	S* (29%)

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.017***	0.007	0.028**	0.007	NS
DIEPAMHR + POM	0.066	0.005	0.060	0.003	NS
Low EC Control @ 203.1 uS/cm + POM	0.057	0.010	0.049	0.011	NS
Confluence of Lindsey Sl. and Cache Sl.	0.091	0.012	0.076	0.015	NS
Sacramento River at tip of Grand Island (711)	0.073	0.008	0.088	0.005	NS
San Joaquin River at Potato Slough (815)	0.083	0.010	0.090	0.008	NS
Upper Cache Slough at mouth of Ulatis Creek	0.097	0.007	0.094	0.008	NS
Sacramento R. Deep Water Channel, Light 55	0.096	0.005	0.085	0.012	NS
Old River at mouth of Holland Cut (915)	0.089	0.008	0.042	0.011	S* (47%)
Old River, western arm at railroad bridge (902)[4]	0.092	0.005	0.028	0.008	-

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

***: $P < 0.001$

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

3. Survival in sample 915 and 902 treated with PBO was reduced compared to the DIEPAMHR without POM, but not compared to the DIEPAMHR + POM.

4. Weight of 902 + PBO could not be examined statistically because only 2 replicates had surviving animals.

Table B 40-2. Summary of 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 8/28/08.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Confluence of Lindsey Sl. And Cache Sl.	209	25.0	7.46	8.2	40.8	0.08	0.001
Sacramento River at tip of Grand Island (711)	201	24.5	7.29	7.2	6.8	0.27	0.003
San Joaquin River at Potato Slough (815)	284	24.9	7.39	7.6	3.9	0.07	0.001
Upper Cache Slough at mouth of Ulatis Creek	287	22.8	7.92	8.4	53.1	0.06	0.002
Sacramento R. Deep Water Channel, Light 55	294	23.6	7.17	8.1	53.5	0.09	0.001
Old River at mouth of Holland Cut (915)	650	25.1	7.33	7.8	6.5	0.04	0.000
Old River, western arm at railroad bridge (902)	741	24.9	7.52	8.2	5.3	0.02	0.000

Table B 40-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 8/29/08 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 8/28/08.

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			
DIEPAMH	345	23.0	24.5	6.5	8.4	7.51	8.29	104	60	-
DIEPAMH + POM	352	23.8	24.5	6.4	8.3	7.48	8.27	104	60	-
Low EC Control @ 203.1 uS/cm + POM	225	24.2	24.5	6.8	8.8	7.33	8.28	58	39	-
Confluence of Lindsey Sl. and Cache Sl.	226	24.4	24.5	6.8	8.7	7.61	8.17	72	82	0.006
Sacramento River at tip of Grand Island (711)	217	24.5	24.6	6.5	8.7	7.56	7.91	72	82	0.011
San Joaquin River at Potato Slough (815)	321	24.6	24.6	6.8	8.7	7.59	8.03	76	82	0.004
Upper Cache Slough at mouth of Ulatis Creek	244	24.5	24.6	6.7	8.5	7.68	8.10	76	88	0.004
Sacramento R. Deep Water Channel, Light 55	320	24.7	24.7	7.0	8.6	7.67	8.27	84	90	0.008
Old River at mouth of Holland Cut (915)	656	24.6	24.9	6.7	8.8	7.59	8.02	104	72	0.002
Old River, western arm at railroad bridge (902)	744	24.6	25.1	6.9	8.7	7.57	8.08	112	86	0.001
DIEPAMH + 25 ppb PBO	365	24.8	25.1	6.7	8.5	7.52	8.25	-	-	-
DIEPAMH + POM + 25 ppb PBO	371	24.7	25.0	6.8	8.4	7.46	8.25	-	-	-
Low EC Control @ 203.1 uS/cm + POM + 25 ppb PBO	232	24.7	25.2	6.6	8.9	7.33	8.17	-	-	-
Confluence of Lindsey Sl. and Cache Sl. + 25 ppb PBO	242	24.5	25.2	6.8	8.6	7.62	8.13	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	232	24.7	25.2	6.9	8.9	7.55	7.92	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	322	24.8	25.1	6.9	8.7	7.59	8.00	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25	262	24.8	25.0	7.2	8.9	7.60	8.08	-	-	-

ppb PBO										
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	322	24.7	25.1	6.8	8.8	7.69	8.17	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	670	24.8	25.2	6.9	8.7	7.61	8.04	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	746	24.8	25.3	6.9	8.9	7.52	8.15	-	-	-

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 B 41-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 9/10/2008 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 8/26/2008 - 8/27/2008.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	100	0.0	95	2.9	NS
DIEPAMHR + POM	95	5.0	90	10.0	NS
Low EC @194.4 uS/cm + POM	90	4.1	100	0.0	NS
High EC @ 14.79 mS/cm + POM	100	0.0	71	14.1	NS
High EC @ 19.04 mS/cm + POM	100	0.0	73	2.5	S* (73%)
Sacramento River at Hood DWR Station[3]	100	0.0	82	17.9	NS
Rough and Ready DWR station, Stockton	100	0.0	97	2.8	NS
Suisun Slough at Rush Ranch[4]	100	0.0	100	0.0	NS
Suisun Bay off Chipps Island (508)	100	0.0	89	7.9	NS
Montezuma Slough at Nurse Slough (609)[4]	100	0.0	100	0.0	NS
Grizzly Bay at Dolphin (602)[5]	100	0.0	90	4.1	NS
Field Dup.: Montezuma Slough at Nurse Slough (609)[4]	100	0.0	-	-	NS
Trip Blank: DIEPAMHR	98	2.5	-	-	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.046	0.002	0.043	0.005	NS
DIEPAMHR + POM	0.048	0.004	0.038	0.005	NS
Low EC @194.4 uS/cm + POM	0.050	0.010	0.040	0.007	NS
High EC @ 14.79 mS/cm + POM	0.051	0.009	0.038	0.005	NS
High EC @ 19.04 mS/cm + POM	0.051	0.005	0.042	0.005	NS
Sacramento River at Hood DWR Station[3]	0.061	0.010	0.056	0.017	NS
Rough and Ready DWR station, Stockton	0.083	0.003	0.063	0.005	S* (76%)
Suisun Slough at Rush Ranch[4]	0.054	0.005	0.037	0.002	S* (69%)
Suisun Bay off Chipps Island (508)	0.063	0.003	0.049	0.004	S* (78%)

Montezuma Slough at Nurse Slough (609)[4]	0.055	0.005	0.053	0.005	NS
Grizzly Bay at Dolphin (602)[5]	0.054	0.004	0.045	0.007	NS
Field Dup.: Montezuma Slough at Nurse Slough (609)[4]	0.061	0.008	-	-	NS
Trip Blank: DIEPAMHR	0.052	0.003	-	-	NS

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

Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.
3. This low conductivity sample was compared to the Low EC Control.
4. This high conductivity sample was compared to the High EC Control @ 14.79 mS/cm.
5. This high conductivity sample was compared to the High EC Control @ 19.04 mS/cm.

Table B 41-2. Summary of 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 8/26/08 - 8/27/08.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (^{o}C)	pH	DO (mg/L)			
Sacramento River at Hood DWR Station	282	27.7	7.45	7.1	19.4	0.24	0.004
Rough and Ready DWR station, Stockton	845	29.8	7.58	6.3	13.4	0.06	0.002
Suisun Slough at Rush Ranch	14330	23.1	7.53	6.7	20.6	0.07	0.001
Suisun Bay off Chipps Island (508)	6490	22.2	7.80	8.2	7.3	0.01	0.000
Montezuma Slough at Nurse Slough (609)	13440	23.0	7.50	7.4	23.8	0.02	0.000
Grizzly Bay at Dolphin (602)	18280	21.8	7.71	8.0	16.7	0.05	0.001
Field Dup.: Montezuma Slough at Nurse Slough (609)	13440	23.0	7.50	7.4	23.8	0.04	0.000
Trip Blank: DIEPAMHR	350	21.9	8.00	8.0	0.2	0.00	0.000

Table B 41-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 9/10/08 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 8/26/08 - 8/27/08.

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	23.1	23.8	7.3	8.3	7.69	8.00	104	56	-
DIEPAMHR + POM	357	22.7	23.8	7.2	8.3	7.61	8.13	104	56	-
Low EC @194.4 uS/cm + POM	205	23.5	23.7	7.3	8.8	7.41	7.93	64	36	-
High EC @ 14.79 mS/cm + POM	14475	23.0	23.8	7.2	8.8	7.65	8.03	2400	83	-
High EC @ 19.04 mS/cm + POM	18050	23.3	23.8	7.0	8.9	7.68	8.02	1560	90	-
Sacramento River at Hood DWR Station	200	24.0	24.0	6.9	8.8	7.72	8.12	72	80	0.015
Rough and Ready DWR station, Stockton	862	23.1	24.0	7.0	8.9	7.92	8.14	160	98	0.003
Suisun Slough at Rush Ranch	13415	23.4	24.0	7.0	8.7	7.89	8.14	1680	168	0.002
Suisun Bay off Chipps Island (508)	6120	23.7	23.9	7.2	8.9	7.69	7.97	600	82	0.000
Montezuma Slough at Nurse Slough (609)	12720	23.5	24.0	7.0	8.3	7.72	8.00	1720	111	0.001
Grizzly Bay at Dolphin (602)	17185	23.9	24.9	6.9	8.3	7.68	7.82	1920	95	0.001
Field Dup.: Montezuma Slough at Nurse Slough (609)	13095	23.7	24.0	6.7	8.6	7.77	7.90	1560	111	0.001
Trip Blank: DIEPAMHR	649	23.8	24.0	7.1	8.9	7.69	8.14	104	62	0.000
DIEPAMHR + 25 ppb PBO	387	23.4	23.7	7.2	8.2	7.66	8.13	-	-	-
DIEPAMHR + POM + 25 ppb PBO	438	23.3	25.3	7.0	8.2	7.65	8.12	-	-	-
Low EC @194.4 uS/cm + POM + 25 ppb PBO	242	23.3	23.9	7.1	8.6	7.50	7.92	-	-	-
High EC @ 14.79 mS/cm + POM + 25 ppb PBO	14330	23.3	23.9	7.0	8.5	7.53	8.01	-	-	-
High EC @ 19.04 mS/cm + POM + 25 ppb PBO	18265	23.1	23.8	7.1	8.9	7.65	8.07	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	192	23.3	24.4	6.5	8.6	7.73	8.87	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	740	23.2	24.1	7.1	8.6	7.89	8.25	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	13095	23.1	25.3	6.7	8.6	7.91	8.12	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	6195	23.3	24.4	7.1	8.4	7.72	7.94	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	13110	23.2	24.1	6.8	8.4	7.77	7.92	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	16995	23.2	24.5	7.3	8.4	7.63	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 B 42-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 9/11/2008 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 9/09/2008 - 9/10/2008.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	98	2.5	95	5.0	NS
DIEPAMHR + POM	90	4.1	95	3.1	NS
Low EC @ 200.7 uS/cm + POM	94	3.3	100	0.0	NS
High EC @ 15.94 mS/cm + POM	100	0.0	98	2.5	NS
High EC @ 21.87 mS/cm + POM	100	0.0	98	2.5	NS
Sacramento River at Hood DWR Station[3]	98	2.5	100	0.0	NS
Rough and Ready DWR station, Stockton	100	0.0	98	2.5	NS
Suisun Slough at Rush Ranch[4]	100	0.0	100	0.0	NS
Suisun Bay off Chipps Island (508)	100	0.0	98	2.5	NS
Montezuma Slough at Nurse Slough (609)[4]	100	0.0	100	0.0	NS
Grizzly Bay at Dolphin (602)[5]	95	5.0	95	5.0	NS
Field Dup.: Montezuma Slough at Nurse Slough (609)[4]	98	2.5	-	-	NS
Field Dup.: Rough and Ready DWR station, Stockton	100	0.0	-	-	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.046	0.006	0.073	0.009	S* (159%)
DIEPAMHR + POM	0.062	0.007	0.051	0.013	NS
Low EC @ 200.7 uS/cm + POM	0.047	0.004	0.054	0.006	NS
High EC @ 15.94 mS/cm + POM	0.046	0.002	0.050	0.008	NS
High EC @ 21.87 mS/cm + POM	0.046*	0.005	0.022	0.002	S* (48%)
Sacramento River at Hood DWR Station[3]	0.096	0.010	0.079	0.010	NS
Rough and Ready DWR station, Stockton	0.118	0.012	0.093	0.008	NS
Suisun Slough at Rush Ranch[4]	0.053	0.003	0.059	0.005	NS
Suisun Bay off Chipps Island (508)	0.091	0.010	0.057	0.005	S* (63%)
Montezuma Slough at Nurse Slough (609)[4]	0.072	0.005	0.036	0.009	S* (50%)
Grizzly Bay at Dolphin (602)[5]	0.056	0.003	0.027	0.009	S* (48%)
Field Dup.: Montezuma Slough at Nurse Slough (609)[4]	0.071	0.004	-	-	NS
Field Dup.: Rough and Ready DWR station, Stockton	0.097	0.006	-	-	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

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

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

4. This high conductivity sample was compared to the High EC Control @ 15.94 mS/cm.

5. This high conductivity sample was compared to the High EC Control @ 21.87 mS/cm.

Table B 42-2. Summary of 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 9/09/08 - 9/10/08.

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 Hood DWR Station	202	22.4	7.52	7.5	9.7	0.23	0.003
Rough and Ready DWR station, Stockton	710	25.4	7.61	6.4	10.2	0.03	0.001
Suisun Slough at Rush Ranch	15130	24.3	7.54	7.3	28.6	0.08	0.001
Suisun Bay off Chipps Island (508)	8010	20.4	7.86	8.5	8.1	0.03	0.001
Montezuma Slough at Nurse Slough (609)	14850	21.3	7.70	7.7	16.6	0.02	0.000
Grizzly Bay at Dolphin (602)	20520	20.2	7.93	8.0	9.4	0.00	0.000
Field Dup.: Montezuma Slough at Nurse Slough (609)	14850	21.3	7.70	7.7	17.3	0.02	0.000
Field Dup.: Rough and Ready DWR station, Stockton	710	25.4	7.61	6.4	11.7	0.06	0.001

Table B 42-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 9/11/08 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 9/09/08 - 9/10/08.

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 (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	327	21.9	23.4	7.4	8.2	7.69	8.08	104	56	-
DIEPAMHR + POM	334	21.8	23.3	7.5	8.2	7.73	8.08	104	56	-
Low EC @ 200.7 uS/cm + POM	193	21.8	23.4	7.2	8.6	7.49	7.90	56	36	-
High EC @ 15.94 mS/cm + POM	15140	21.8	23.3	7.2	8.1	7.58	7.98	1800	82	-
High EC @ 21.87 mS/cm + POM	20545	21.8	23.4	7.1	7.9	7.68	7.98	2520	90	-
Sacramento River at Hood DWR Station	199	21.8	23.4	7.0	8.4	7.74	7.98	48	82	0.009
Rough and Ready DWR station, Stockton	682	21.8	23.3	7.3	8.3	7.87	8.08	156	106	0.001
Suisun Slough at Rush Ranch	14185	21.7	23.3	6.9	8.2	7.75	8.10	2000	176	0.002
Suisun Bay off Chipps Island (508)	7765	21.8	23.4	7.2	8.1	7.68	7.82	920	84	0.001
Montezuma Slough at Nurse Slough (609)	14215	21.8	23.4	7.1	8.2	7.69	7.88	1680	110	0.000
Grizzly Bay at Dolphin (602)	19380	21.8	23.5	7.3	8.2	7.65	7.82	2320	92	0.000
Field Dup.: Montezuma Slough at Nurse Slough (609)	14195	21.7	23.6	7.1	8.2	7.69	7.90	1720	112	0.000
Field Dup.: Rough and Ready DWR station, Stockton	696	21.7	23.3	7.4	8.2	7.83	8.12	152	106	0.002
DIEPAMHR + 25 ppb PBO	344	21.7	23.0	7.5	8.1	7.67	8.09	-	-	-
DIEPAMHR + POM + 25 ppb PBO	345	21.7	23.0	7.4	8.3	7.73	8.09	-	-	-
Low EC @ 200.7 uS/cm + POM + 25 ppb PBO	199	21.8	22.9	7.4	8.5	7.51	7.90	-	-	-
High EC @ 15.94 mS/cm + POM + 25 ppb PBO	14950	21.8	23.1	7.2	8.1	7.59	7.97	-	-	-
High EC @ 21.87 mS/cm + POM + 25 ppb PBO	20575	21.8	23.2	7.2	8.1	7.70	7.98	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	205	21.8	23.3	7.0	8.3	7.76	8.00	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	693	22.2	23.3	7.3	8.5	7.87	8.07	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	14190	22.0	23.7	7.1	8.2	7.73	8.12	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	7770	22.0	23.6	7.3	8.2	7.70	7.87	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	14230	22.1	23.6	7.1	8.3	7.74	7.93	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	19455	22.6	23.6	7.3	8.2	7.65	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 B 43-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 9/12/08 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 9/11/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	98	2.5	87	4.7	NS
Low EC Control @ 209.7 uS/cm	95	2.9	100	0.0	NS
Confluence of Lindsey Sl. and Cache Sl.[3]	100	0.0	100	0.0	NS
Sacramento River at tip of Grand Island (711)[3]	98	2.5	100	0.0	NS
San Joaquin River at Potato Slough (815)	100	0.0	98	2.5	NS
Upper Cache Slough at mouth of Ulatis Creek[3]	98	2.5	95	4.5	NS
Sacramento R. Deep Water Channel, Light 55	98	2.5	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)	100	0.0	100	0.0	NS
Bottle Blank: DIEPAMHR	98	2.5	-	-	NA
Trip Blank: DIEPAMHR	95	2.8	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.065	0.006	0.077	0.003	NS
Low EC Control @ 209.7 uS/cm	0.041*	0.007	0.068	0.006	S* (166%)
Confluence of Lindsey Sl. and Cache Sl.[3]	0.106	0.011	0.123	0.006	NS
Sacramento River at tip of Grand Island (711)[3]	0.109	0.003	0.120	0.010	NS
San Joaquin River at Potato Slough (815)	0.145	0.007	0.128	0.008	NS
Upper Cache Slough at mouth of Ulatis Creek[3]	0.121	0.006	0.118	0.004	NS
Sacramento R. Deep Water Channel, Light 55	0.125	0.006	0.120	0.015	NS
Old River at mouth of Holland Cut (915)	0.135	0.007	0.132	0.011	NS
Old River, western arm at railroad bridge (902)	0.153	0.010	0.127	0.007	NS
Bottle Blank: DIEPAMHR	0.080	0.001	-	-	NA
Trip Blank: DIEPAMHR	0.068	0.004	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

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

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

Table B 43-2. Summary of 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 9/11/08.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (^{o}C)	pH	DO (mg/L)			
Confluence of Lindsey Sl. and Cache Sl.	225	20.5	7.74	8.2	42.7	0.11	0.002
Sacramento River at tip of Grand Island (711)	330	23.5	7.28	7.8	5.4	0.48	0.004
San Joaquin River at Potato Slough (815)	449	22.4	7.63	8.1	3.5	0.07	0.001
Upper Cache Slough at mouth of Ulatis Creek	345	22.0	7.70	8.9	50.3	0.05	0.001
Sacramento R. Deep Water Channel, Light 55	309	21.6	7.83	8.2	29.7	0.03	0.001
Old River at mouth of Holland Cut (915)	638	23.5	7.71	7.5	5.0	0.07	0.002
Old River, western arm at railroad bridge (902)	1093	23.8	7.36	8.4	5.4	0.03	0.000
Trip Blank: DIEPAMHR	345	22.3	8.11	8.2	0.6	0.01	0.001

Table B 43-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 9/12/08 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 9/11/08.

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 (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	340	21.9	23.2	7.2	8.2	7.73	8.14	104	56	-
Low EC Control @ 209.7 uS/cm	219	21.9	23.2	7.3	8.6	7.52	7.90	56	82	-
Confluence of Lindsey Sl. and Cache Sl.	217	21.9	23.3	7.0	8.9	7.81	8.11	72	86	0.006
Sacramento River at tip of Grand Island (711)	210	21.9	23.4	6.9	8.8	7.74	7.97	68	84	0.016
San Joaquin River at Potato Slough (815)	453	21.9	23.3	7.0	8.4	7.83	8.05	92	82	0.003
Upper Cache Slough at mouth of Ulatis Creek	235	21.8	23.2	7.2	8.3	7.93	8.15	80	90	0.003
Sacramento R. Deep Water Channel, Light 55	301	21.8	23.2	7.3	8.4	7.97	8.21	92	92	0.002
Old River at mouth of Holland Cut (915)	623	21.8	23.3	7.1	8.5	7.80	8.08	108	80	0.003
Old River, western arm at railroad bridge (902)	916	21.8	23.3	7.3	8.9	7.80	8.08	140	80	0.002
Bottle Blank: DIEPAMHR	352	21.9	23.4	7.1	8.6	7.80	8.15	104	60	0.001
Trip Blank: DIEPAMHR	353	21.8	23.3	7.4	8.5	7.77	8.15	108	62	0.001
DIEPAMHR + 25 ppb PBO	339	21.8	23.5	7.1	8.1	7.74	8.16	-	-	-
Low EC Control @ 209.7 uS/cm + 25 ppb PBO	215	21.9	23.5	7.3	8.2	7.52	7.91	-	-	-
Confluence of Lindsey Sl. and Cache Sl. + 25 ppb PBO	217	22.1	23.5	7.2	8.4	7.81	8.16	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	198	22.0	23.5	7.1	8.4	7.74	7.99	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	448	22.2	23.5	7.1	8.3	7.86	8.07	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	220	22.4	23.6	7.3	8.3	7.95	8.17	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	297	22.8	23.4	7.1	8.4	7.98	8.21	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	616	22.6	23.4	7.1	8.3	7.82	8.01	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	926	22.7	23.5	7.2	8.4	7.81	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 B 44-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 9/25/08 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 9/23/08 - 9/24/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	100	0.0	80	20.0	NS
Low EC Control @ 172 µS/cm	97	2.8	100	0.0	NS
High EC Control @ 17.38 mS/cm	72*	20.9	65	22.5	NS
Sacramento River at Hood DWR Station[3]	100	0.0	95	3.1	NS
Rough and Ready DWR station, Stockton	100	0.0	98	2.5	NS
Suisun Slough at Rush Ranch[4]	98	2.5	73	11.9	NS
Suisun Bay off Chipps Island (508)	98	2.5	98	2.5	NS
Montezuma Slough at Nurse Slough (609)[4]	100	0.0	83	5.6	S* (83%)
Grizzly Bay at Dolphin (602)[4]	98	2.5	93	4.8	NS
Trip Blank: DIEPAMHR	95	3.1	-	-	NA
Field Dup.: Sacramento River at Hood DWR Station[3]	95	3.1	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.064	0.004	0.063	0.005	NS
Low EC Control @ 172 µS/cm	0.060	0.006	0.064	0.005	NS
High EC Control @ 17.38 mS/cm	0.055	0.003	0.043*	0.003	S* (78%)
Sacramento River at Hood DWR Station[3]	0.078	0.001	0.079	0.006	NS
Rough and Ready DWR station, Stockton	0.132	0.016	0.084	0.010	S* (64%)
Suisun Slough at Rush Ranch[4]	0.048	0.009	0.041	0.009	NS
Suisun Bay off Chipps Island (508)	0.086	0.009	0.076	0.001	NS
Montezuma Slough at Nurse Slough (609)[4]	0.046	0.005	0.062	0.011	NS
Grizzly Bay at Dolphin (602)[4]	0.061	0.009	0.068	0.006	NS
Trip Blank: DIEPAMHR	0.062	0.005	-	-	NA
Field Dup.: Sacramento River at Hood DWR Station[3]	0.079	0.008	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

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

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

4. This high conductivity sample was compared to the High EC Control.

Table B 44-2. Summary of 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 9/23/08 - 9/24/08.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (^{o}C)	pH	DO (mg/L)			
Sacramento River at Hood DWR Station	172	20.8	7.25	7.6	6.7	0.29	0.002
Rough and Ready DWR station, Stockton	724	23.3	7.24	7.8	12.9	0.07	0.001
Suisun Slough at Rush Ranch	16070	20.9	7.31	7.8	15.2	0.06	0.000
Suisun Bay off Chipps Island (508)	5150	19.2	7.59	9.0	9.4	0.05	0.001
Montezuma Slough at Nurse Slough (609)	15420	19.9	7.33	7.9	14.9	0.01	0.000
Grizzly Bay at Dolphin (602)	16680	18.9	7.77	8.8	17.5	0.06	0.001
Trip Blank: DIEPAMHR	342	20.6	8.05	8.6	0.3	0.00	0.000
Field Dup.: Sacramento River at Hood DWR Station	172	20.8	7.25	7.6	6.8	0.27	0.002

Table B 44-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 9/25/08 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 9/23/08 - 9/24/08.

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	21.8	24.0	7.4	8.0	7.49	8.00	104	56	-
Low EC Control @ 172 µS/cm	166	21.9	23.9	7.7	8.4	7.35	7.75	52	30	-
High EC Control @ 17.38 mS/cm	16395	21.8	23.8	7.5	8.7	7.66	7.93	1920	88	-
Sacramento River at Hood DWR Station	169	21.9	23.7	7.0	8.4	7.54	7.79	60	68	0.008
Rough and Ready DWR station, Stockton	696	21.9	24.0	7.1	8.6	7.87	8.04	152	106	0.003
Suisun Slough at Rush Ranch	15275	22.1	23.9	7.1	8.5	7.70	8.07	2320	186	0.001
Suisun Bay off Chipps Island (508)	4940	22.0	24.0	7.2	8.9	7.73	7.88	572	88	0.001
Montezuma Slough at Nurse Slough (609)	14725	22.4	24.0	6.9	8.6	7.71	7.88	1880	110	0.000
Grizzly Bay at Dolphin (602)	16370	22.8	23.8	7.1	8.3	7.67	7.77	2000	92	0.001
Trip Blank: DIEPAMHR	350	22.6	23.7	7.7	8.7	7.72	8.04	104	62	0.000
Field Dup.: Sacramento River at Hood DWR Station	174	22.7	24.0	7.2	8.6	7.62	7.94	60	68	0.009
DIEPAMHR + 25 ppb PBO	343	22.4	23.2	7.7	8.3	7.73	8.02	-	-	-
Low EC Control @ 172 µS + 25 ppb PBO	173	22.0	23.1	6.9	8.6	7.37	7.78	-	-	-
High EC Control @ 17.38 mS + 25 ppb PBO	16185	22.6	22.9	7.2	8.4	7.68	7.93	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	204	22.2	22.9	7.1	8.4	7.65	7.88	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	692	22.1	23.0	7.1	8.5	7.81	8.10	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	15280	22.2	22.9	7.0	8.2	7.71	8.05	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	5030	22.7	23.3	7.2	8.4	7.71	7.88	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	14770	22.3	23.3	7.4	8.5	7.79	7.89	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	16130	22.7	23.7	7.1	8.5	7.70	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 B 45-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 09/26/08 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 09/25/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	92	2.8	92	4.8	NS
Low EC Control @ 175 uS/cm	95	2.9	91	5.9	NS
Sacramento River at tip of Grand Island (711)[3]	87	3.1	95	2.9	NS
Old River at mouth of Holland Cut (915)	100	0.0	100	0.0	NS
Upper Cache Slough at mouth of Ulatis Creek	100	0.0	100	0.0	NS
Old River, western arm at railroad bridge (902)	100	0.0	100	0.0	NS
Confluence of Lindsey Sl. And Cache Sl.[3]	95	5.0	95	2.9	NS
San Joaquin River at Potato Slough (815)	100	0.0	98	2.5	NS
Sacramento R. Deep Water Channel, Light 55	97	2.8	90	4.1	NS
Field Dup.: Old River at mouth of Holland Cut (915)	100	0.0	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.043	0.009	0.031	0.007	NS
Low EC Control @ 175 uS/cm	0.046	0.002	0.034	0.012	NS
Sacramento River at tip of Grand Island (711)[3]	0.049	0.014	0.061	0.007	NS
Old River at mouth of Holland Cut (915)	0.077	0.004	0.078	0.004	NS
Upper Cache Slough at mouth of Ulatis Creek	0.067	0.009	0.092	0.018	NS
Old River, western arm at railroad bridge (902)	0.086	0.004	0.084	0.014	NS
Confluence of Lindsey Sl. And Cache Sl.[3]	0.065	0.009	0.069	0.006	NS
San Joaquin River at Potato Slough (815)	0.051	0.006	0.058	0.005	NS
Sacramento R. Deep Water Channel, Light 55	0.076	0.006	0.068	0.005	NS
Field Dup.: Old River at mouth of Holland Cut (915)	0.077	0.008	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

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

3. These low conductivity samples were compared to the Low EC Control.

Table B 45-2. Summary of 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 09/25/2008.

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)	176	21.2	7.29	7.9	4.0	0.43	0.003
Old River at mouth of Holland Cut (915)	704	21.5	7.51	8.6	7.3	0.03	0.000
Upper Cache Slough at mouth of Ulatis Creek	287	19.7	7.83	8.5	48.0	0.02	0.000
Old River, western arm at railroad bridge (902)	704	21.7	7.45	8.9	6.7	0.00	0.000
Confluence of Lindsey Sl. And Cache Sl.	220	20.1	7.56	8.4	37.1	0.07	0.001
San Joaquin River at Potato Slough (815)	294	21.1	7.43	8.4	4.5	0.06	0.001
Sacramento R. Deep Water Channel, Light 55	315	20.0	7.45	8.5	51.9	0.00	0.000
Field Dup. of Old River at mouth of Holland Cut (915)	704	21.5	7.51	8.6	7.1	0.01	0.000

Table B 45-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 09/26/2008 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 09/25/2008.

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	22.0	24.1	7.2	8.3	7.55	8.15	104	56	-
Low EC Control @ 175 uS/cm	217	22.9	24.1	7.4	8.8	7.35	7.99	60	32	-
Sacramento River at tip of Grand Island (711)	201	22.0	23.5	7.0	8.7	7.61	7.89	60	68	0.015
Old River at mouth of Holland Cut (915)	685	21.9	23.7	7.3	8.8	7.68	8.03	116	80	0.001
Upper Cache Slough at mouth of Ulatis Creek	274	21.9	23.5	7.3	8.6	7.78	8.17	88	98	0.001
Old River, western arm at railroad bridge (902)	680	21.9	23.6	7.1	8.6	7.66	8.10	116	78	0.000
Confluence of Lindsey Sl. And Cache Sl.	232	21.9	23.9	7.4	8.6	7.74	8.11	100	90	0.004
San Joaquin River at Potato Slough (815)	302	22.0	23.9	7.1	8.6	7.67	8.08	64	78	0.003
Sacramento R. Deep Water Channel, Light 55	327	22.0	23.7	7.1	8.8	7.74	8.17	104	96	0.000
Field Dup. of Old River at mouth of Holland Cut (915)	679	21.8	23.8	7.5	8.7	7.68	8.08	116	82	0.001
DIEPAMHR + 25 ppb PBO	343	21.8	23.3	7.3	8.4	7.53	8.16	-	-	-
Low EC Control @ 175 uS/cm + 25 ppb PBO	194	21.9	23.3	7.4	8.6	7.38	7.96	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	191	21.9	23.9	7.0	8.7	7.60	7.97	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	677	21.9	23.8	7.3	8.8	7.70	8.10	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	271	21.9	23.8	7.3	8.6	7.83	8.14	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	684	21.9	23.7	7.2	8.6	7.69	8.11	-	-	-
Confluence of Lindsey Sl. And Cache Sl. + 25 ppb PBO	235	22.0	23.7	7.3	8.6	7.78	8.14	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	304	22.0	23.5	7.3	8.7	7.70	8.10	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	328	22.1	24.2	7.1	8.6	7.72	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 B 46-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 10/9/08 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 10/7/08 - 10/8/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	100	0.0	100	0.0	NS
Low EC Control @ 164 µS/cm	98	2.5	98	2.5	NS
High EC Control @ 18.42 mS/cm	100	0.0	77*	8.5	NS
Sacramento River at Hood DWR Station[3]	93	4.4	98	2.3	NS
Rough and Ready DWR station, Stockton	100	0.0	100	0.0	NS
Suisun Slough at Rush Ranch[4]	98	2.5	95	2.8	NS
Suisun Bay off Chipps Island (508)	100	0.0	95	5.0	NS
Montezuma Slough at Nurse Slough (609)	100	0.0	100	0.0	NS
Grizzly Bay at Dolphin (602)[4]	95	2.9	86	10.5	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.080	0.012	0.053	0.005	NS
Low EC Control @ 164 µS/cm	0.041*	0.009	0.059	0.009	NS
High EC Control @ 18.42 mS/cm	0.010**	0.002	0.022**	0.004	S* (220%)
Sacramento River at Hood DWR Station[3]	0.022	0.005	0.050	0.006	S* (227%)
Rough and Ready DWR station, Stockton	0.046	0.015	0.075	0.008	NS
Suisun Slough at Rush Ranch[4]	0.037	0.005	0.040	0.006	NS
Suisun Bay off Chipps Island (508)	0.047*	0.008	0.048	0.005	NS
Montezuma Slough at Nurse Slough (609)	0.043*	0.012	0.022**	0.005	NS
Grizzly Bay at Dolphin (602)[4]	0.055	0.003	0.042	0.011	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

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

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

4. This high conductivity sample was compared to the High EC Control.

Table B 46-2. Summary of 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 10/07/08 - 10/08/08.

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 Hood DWR Station	152	22.1	7.42	9.7	5.4	0.32	0.004
Rough and Ready DWR station, Stockton	730	25.0	7.77	9.2	7.0	0.08	0.002
Suisun Slough at Rush Ranch	16020	24.5	7.68	11.0	22.2	0.09	0.002
Suisun Bay off Chipps Island (508)	8060	21.1	7.85	9.2	6.9	0.07	0.002
Montezuma Slough at Nurse Slough (609)	9330	21.4	7.71	9.2	10.8	0.08	0.001
Grizzly Bay at Dolphin (602)	17520	20.3	7.86	9.8	10.1	0.07	0.001

Table B 46-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 10/09/08 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 10/07/08 - 10/08/08.

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	21.5	23.5	7.4	8.4	7.06	8.04	108	60	-
Low EC Control @ 164 µS/cm	162	21.5	23.6	7.8	8.9	7.46	7.84	52	34	-
High EC Control @ 18.42 mS/cm	10934	21.5	23.8	7.4	8.2	7.78	7.90	2560	94	-
Sacramento River at Hood DWR Station	156	21.4	24.1	7.1	8.7	7.65	7.80	56	66	0.007
Rough and Ready DWR station, Stockton	706	21.5	24.0	7.3	8.4	7.83	8.19	160	116	0.002
Suisun Slough at Rush Ranch	15730	21.3	24.2	7.0	8.3	7.64	8.13	1960	176	0.001
Suisun Bay off Chipps Island (508)	7605	21.2	24.2	7.3	8.6	7.71	7.93	880	88	0.002
Montezuma Slough at Nurse Slough (609)	8755	20.9	24.2	7.4	8.5	7.75	7.88	1080	88	0.002
Grizzly Bay at Dolphin (602)	16035	20.5	24.1	7.2	8.2	7.76	8.11	2040	96	0.001
DIEPAMHR + 25 ppb PBO	363	20.9	23.2	7.7	8.5	7.83	8.11	-	-	-
Low EC Control @ 164 µS/cm + 25 ppb PBO	169	20.9	23.1	7.7	8.9	7.46	7.87	-	-	-
High EC Control @ 18.42 mS/cm + 25 ppb PBO	20790	20.8	23.3	7.3	8.1	7.75	7.92	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	155	20.7	23.6	7.5	8.4	7.60	7.83	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	699	20.6	23.4	7.4	8.6	7.84	8.14	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	15205	20.5	23.4	7.1	8.2	7.66	8.14	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	7550	20.9	23.8	7.2	8.4	7.84	7.90	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	8480	20.4	23.7	7.4	8.3	7.78	7.88	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	16645	20.4	23.8	7.0	8.6	7.78	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 B 47-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 10/10/08 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 10/9/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	98	2.5	98	2.5	NS
Low EC Control @ 175 µS/cm	95	5.0	93	2.5	NS
Sacramento River at tip of Grand Island (711)[3]	100	0.0	98	2.3	NS
Upper Cache Slough at mouth of Ulatis Creek	100	0.0	100	0.0	NS
Confluence of Lindsey Sl. And Cache Sl.[3]	100	0.0	100	0.0	NS
San Joaquin River at Potato Slough (815)	100	0.0	78	19.3	NS
Sacramento R. Deep Water Channel, Light 55	98	2.5	100	0.0	NS
Field Dup.: Sacramento R. Deep Water Channel, Light 55	98	2.5	-	-	NA
Field Dup.: Confluence of Lindsey Sl. And Cache Sl.[3]	100	0.0	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.040	0.004	0.037	0.007	NS
Low EC Control @ 175 µS/cm	0.034	0.005	0.037	0.004	NS
Sacramento River at tip of Grand Island (711)[3]	0.060	0.006	0.058	0.004	NS
Upper Cache Slough at mouth of Ulatis Creek	0.060	0.009	0.048	0.010	NS
Confluence of Lindsey Sl. And Cache Sl.[3]	0.073	0.006	0.052	0.009	NS
San Joaquin River at Potato Slough (815)	0.060	0.008	0.068	0.009	NS
Sacramento R. Deep Water Channel, Light 55	0.076	0.011	0.069	0.005	NS
Field Dup.: Sacramento R. Deep Water Channel, Light 55	0.066	0.002	-	-	NA
Field Dup.: Confluence of Lindsey Sl. And Cache Sl.[3]	0.072	0.006	-	-	NA

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

Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

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

3. These low conductivity samples were compared to the Low EC Control.

Table B 47-2. Summary of 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 10/9/08.

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)	175	19.5	7.67	8.7	8.4	0.27	0.004
Upper Cache Slough at mouth of Ulatis Creek	276	18.5	7.75	9.2	40.6	0.05	0.001
Confluence of Lindsey Sl. And Cache Sl.	188	19.6	7.74	8.6	20.5	0.18	0.004
San Joaquin River at Potato Slough (815)	296	20.2	7.75	9.1	5.2	0.10	0.002
Sacramento R. Deep Water Channel, Light 55	265	19.7	7.83	8.8	32.4	0.06	0.001
Field Dup.: Sacramento R. Deep Water Channel, Light 55	265	19.7	7.83	8.8	32.8	0.08	0.002
Field Dup.: Confluence of Lindsey Sl. And Cache Sl.	188	19.6	7.74	8.6	32.8	0.18	0.004

Table B 47-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 10/10/08 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 10/9/08.

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	21.8	23.8	7.8	8.0	7.78	8.07	108	60	-
Low EC Control @ 175 µS/cm	174	21.9	23.6	7.6	8.5	7.52	7.91	52	32	-
Sacramento River at tip of Grand Island (711)	172	21.9	23.5	7.4	8.6	7.67	7.88	60	68	0.010
Upper Cache Slough at mouth of Ulatis Creek	278	22.0	23.8	7.3	8.4	7.95	8.12	96	106	0.003

Confluence of Lindsey Sl. And Cache Sl.	187	21.9	23.6	7.3	8.9	7.72	7.89	68	74	0.006
San Joaquin River at Potato Slough (815)	277	21.8	23.5	7.4	8.5	7.77	7.92	72	78	0.004
Sacramento R. Deep Water Channel, Light 55	261	22.0	23.8	7.1	8.8	7.82	8.06	84	84	0.003
Field Dup.: Sacramento R. Deep Water Channel, Light 55	250	21.9	23.6	7.4	8.6	7.78	8.04	80	86	0.004
Field Dup.: Confluence of Lindsey Sl. And Cache Sl.	185	22.4	23.8	7.3	8.8	7.67	7.89	64	74	-
DIEPAMHR + 25 ppb PBO	337	22.0	22.7	7.6	8.3	7.74	8.06	-	-	-
Low EC Control @ 175 µS/cm + 25 ppb PBO	177	22.0	22.7	7.7	8.7	7.56	7.89	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	173	22.0	22.8	7.5	8.6	7.68	7.94	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	274	21.9	22.6	7.6	8.4	7.94	8.13	-	-	-
Confluence of Lindsey Sl. And Cache Sl. + 25 ppb PBO	186	22.0	22.7	7.2	8.4	7.75	7.89	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	278	22.0	22.6	7.4	8.6	7.75	8.00	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	256	22.1	22.7	7.6	8.5	7.80	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 B 48-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 10/14/08 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 10/14/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	98	2.5	95	5.0	NS
Old River, western arm at railroad bridge (902)	100	0.0	100	0.0	NS
Old River at mouth of Holland Cut (915)	100	0.0	93	2.4	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.053	0.007	0.025	0.007	S* (47%)
Old River, western arm at railroad bridge (902)	0.077	0.006	0.052	0.005	S* (67%)
Old River at mouth of Holland Cut (915)	0.072	0.002	0.058	0.012	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

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

Table B 48-2. Summary of 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 10/14/2008.

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)	527	16.5	7.87	9.7	6.9	0.03	0.001
Old River at mouth of Holland Cut (915)	543	16.4	7.95	9.7	7.2	0.02	0.000

Table B 48-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 10/14/2008 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 10/14/2008.

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	22.7	24.0	7.6	8.3	7.72	8.06	108	60	-
Old River, western arm at railroad bridge (902)	521	23.1	23.7	7.5	8.6	7.79	7.91	100	76	0.001
Old River at mouth of Holland Cut (915)	533	22.8	24.1	7.5	8.5	7.79	7.97	96	78	0.001
DIEPAMHR + 25 ppb PBO	337	23.6	24.2	7.5	8.3	7.71	8.03	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	514	23.2	24.0	7.3	8.6	7.73	7.91	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	530	23.0	24.1	7.5	8.8	7.79	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 B 49-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 10/23/08 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 10/21/08 - 10/22/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	100	0.0	79	9.4	S* (79%)
Low EC Control @ 160.3 uS/cm	100	0.0	93	4.5	NS
High EC Control @ 16.26 mS/cm	100	0.0	69	14.6	S* (69%)
High EC Control @ 21.56 mS/cm	98	2.5	25	25.0	NS
Sacramento River at Hood DWR Station[3]	100	0.0	98	2.5	NS
Rough and Ready DWR station, Stockton	100	0.0	98	2.5	NS
Suisun Slough at Rush Ranch[4]	100	0.0	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
Grizzly Bay at Dolphin (602)[5]	84	15.6	100	0.0	NS
Field Dup: Suisun Slough at Rush Ranch	100	0.0	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.064	0.006	0.013	0.003	S*** (20%)
Low EC Control @ 160.3 uS/cm	0.087	0.005	0.032	0.007	S*** (37%)
High EC Control @ 16.26 mS/cm	0.059	0.005	0.035	0.005	S** (59%)
High EC Control @ 21.56 mS/cm	0.040*	0.006	0.051	-	NA
Sacramento River at Hood DWR Station[3]	0.091	0.005	0.058	0.011	S* (63%)
Rough and Ready DWR station, Stockton	0.122	0.011	0.113	0.007	NS
Suisun Slough at Rush Ranch[4]	0.073	0.008	0.072	0.011	NS
Suisun Bay off Chipps Island (508)	0.098	0.008	0.069	0.003	S* (70%)
Montezuma Slough at Nurse Slough (609)	0.089	0.009	0.074	0.006	NS
Grizzly Bay at Dolphin (602)[5]	0.065	0.011	0.051	0.005	NS
Field Dup: Suisun Slough at Rush Ranch	0.065	0.008	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

***: $P < 0.001$

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

3. This low conductivity sample was compared to the Low EC Control @ 160.3 uS/cm

4. This high conductivity sample was compared to the High EC Control @ 16.26 mS/cm

5. This high conductivity sample was compared to the High EC Control @ 21.56 mS/cm

Table B 49-2. Summary of 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 10/21/08 - 10/22/08.

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 Hood DWR Station	157	18.2	6.80	11.2	4.5	0.37	0.001
Rough and Ready DWR station, Stockton	690	19.0	7.49	8.7	6.6	0.05	0.001
Suisun Slough at Rush Ranch	14890	16.5	7.52	8.3	33.8	0.11	0.001
Suisun Bay off Chipps Island (508)	9730	16.6	7.58	10.1	20.8	0.05	0.000
Montezuma Slough at Nurse Slough (609)	10370	16.5	7.49	8.9	20.5	0.07	0.000

Grizzly Bay at Dolphin (602)	20320	16.4	7.68	9.5	10.9	0.06	0.001
Field Dup: Suisun Slough at Rush Ranch	14890	16.5	7.52	8.3	29.5	0.07	0.000

Table B 49-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 10/23/08 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 10/21/08 - 10/22/08.

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 (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	335	21.5	23.4	7.7	8.5	7.70	8.12	108	60	-
Low EC Control @ 160.3 uS/cm	161	21.6	23.5	7.9	8.7	7.22	7.94	52	30	-
High EC Control @ 16.26 mS/cm	15555	21.5	23.8	7.4	8.2	7.59	7.95	1880	84	-
High EC Control @ 21.56 mS/cm	20385	21.5	24.1	7.3	8.4	7.67	8.06	2560	92	-
Sacramento River at Hood DWR Station	155	21.5	23.9	7.3	8.5	7.62	7.93	52	62	0.015
Rough and Ready DWR station, Stockton	546	21.5	23.9	7.6	8.6	7.80	8.05	128	94	0.003
Suisun Slough at Rush Ranch	14380	21.5	23.9	7.5	8.6	7.83	8.03	1680	162	0.003
Suisun Bay off Chipps Island (508)	9650	21.5	24.0	7.6	8.8	7.64	7.92	1160	90	0.002
Montezuma Slough at Nurse Slough (609)	10060	21.5	23.9	7.5	8.4	7.64	7.90	2320	94	0.002
Grizzly Bay at Dolphin (602)	19650	21.5	23.9	7.3	8.4	7.68	7.88	1120	88	0.001
Field Dup: Suisun Slough at Rush Ranch	14415	21.7	24.0	7.4	8.4	7.83	8.08	1600	160	0.002
DIEPAMHR + 25 ppb PBO	373	21.5	24.1	7.8	8.5	7.62	8.21	-	-	-
Low EC Control @ 160.3 uS/cm + 25 ppb PBO	169	21.4	24.1	7.1	8.7	7.46	7.89	-	-	-
High EC Control @ 16.26 mS/cm + 25 ppb PBO	15675	21.5	24.1	7.6	8.5	7.59	7.95	-	-	-
High EC Control @ 21.56 mS/cm + 25 ppb PBO	20520	21.3	24.0	7.5	8.3	7.66	8.04	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	193	21.3	24.1	7.4	8.8	7.63	7.83	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	562	21.5	24.0	7.6	8.6	7.86	8.28	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	14230	22.5	24.1	7.5	8.3	7.86	8.08	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	9680	22.2	24.0	7.5	8.7	7.64	7.88	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	10015	21.7	24.0	7.3	8.5	7.67	7.84	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	19550	21.5	23.8	7.3	8.4	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 B 50-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 10/24/08 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 10/23/08

Treatment	Survival (%)[1]

	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	100	0.0	100	0.0	NS
Low EC Control at 187 µS/cm	100	0.0	100	0.0	NS
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	98	2.5	NS
Upper Cache Slough at mouth of Ulatis Creek	100	0.0	100	0.0	NS
Confluence of Lindsey Sl. and Cache Sl. [3]	100	0.0	100	0.0	NS
San Joaquin River at Potato Slough (815)	100	0.0	100	0.0	NS
Old River, western arm at railroad bridge (902)	100	0.0	100	0.0	NS
Old River at mouth of Holland Cut (915)	100	0.0	100	0.0	NS
Field Dup: Sacramento River at Tip of Grand Island [3]	100	0.0	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.057	0.003	0.064	0.007	NS
Low EC Control at 187 µS/cm	0.062	0.003	0.060	0.005	NS
Sacramento R. Deep Water Channel, Light 55	0.084	0.006	0.090	0.006	NS
Sacramento River at tip of Grand Island (711)[3]	0.076	0.005	0.091	0.005	NS
Upper Cache Slough at mouth of Ulatis Creek	0.093	0.012	0.090	0.008	NS
Confluence of Lindsey Sl. and Cache Sl. [3]	0.074	0.012	0.073	0.006	NS
San Joaquin River at Potato Slough (815)	0.095	0.007	0.096	0.009	NS
Old River, western arm at railroad bridge (902)	0.087	0.010	0.085	0.007	NS
Old River at mouth of Holland Cut (915)	0.088	0.006	0.093	0.004	NS
Field Dup: Sacramento River at Tip of Grand Island [3]	0.086	0.001	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

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

3. These low conductivity samples were compared to the Low EC Control.

Table B 50-2. Summary of 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 10/23/08

Treatment	Field Chemistry	Turbidity	Total	Unionized

	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)	(NTU)	Ammonia Nitrogen (mg/L)	Ammonia (mg/L)
Sacramento R. Deep Water Channel, Light 55	303	16.4	7.08	10.3	39.5	0.07	0.000
Upper Cache Slough at mouth of Ulatis Cut	266	15.4	7.79	10.4	25.7	0.19	0.000
Confluence of Lindsey Sl. and Cache Slough	190	16.6	7.42	10.0	8.4	0.35	0.001
Sacramento River at tip of Grand Island (711)	165	16.6	7.15	9.5	35.1	0.03	0.001
San Joaquin River at Potato Slough (815)	285	16.5	7.38	10.6	3.3	0.12	0.001
Old River, western arm at railroad bridge (902)	621	17.4	7.50	10.4	6.1	0.03	0.000
Old River at mouth of Holland Cut (915)	531	17.6	7.77	10.7	6.4	0.00	0.000
Field Dup: Sacramento River at tip of Grand Island	164	16.6	7.15	9.5	8.5	0.34	0.001

Table B 50-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 10/24/08 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 10/23/08.

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	21.5	24.1	7.6	8.3	7.78	8.12	108	60	-
Low EC Control at 187 µS/cm	185	21.4	24.0	7.5	8.9	7.53	7.97	56	34	-
Sacramento R. Deep Water Channel, Light 55	288	21.4	24.1	7.3	8.9	7.79	8.09	96	86	0.004
Sacramento River at tip of Grand Island (711)	180	21.4	24.2	7.3	8.8	7.64	8.00	64	68	0.017
Upper Cache Slough at mouth of Ulatis Creek	255	21.4	24.2	7.3	8.7	7.89	8.16	88	92	0.002
Confluence of Lindsey Sl. and Cache Sl.	186	21.4	24.1	6.9	8.7	7.62	8.04	72	70	0.010
San Joaquin River at Potato Slough (815)	274	21.4	24.2	7.4	8.8	7.65	8.05	76	70	0.006
Old River, western arm at railroad bridge (902)	604	21.4	24.1	7.4	8.8	7.80	8.06	112	78	0.002
Old River at mouth of Holland Cut (915)	521	21.4	24.2	7.7	8.8	7.86	8.12	104	76	0.000
Field Dup: Sacramento River at Tip of Grand Island	182	21.4	24.1	7.1	8.6	7.61	7.98	64	66	0.013
DIEPAMHR + 25 ppb PBO	327	21.4	23.9	7.6	8.5	7.77	8.11	-	-	-
Low EC Control at 187 µS/cm + 25 ppb PBO	181	21.4	24.0	7.6	8.5	7.51	7.95	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	292	21.4	24.0	7.1	8.9	7.82	8.13	-	-	-
Sacramento River at tip of Grand Island + 25 ppb pBO	180	21.5	24.1	7.3	8.4	7.63	7.98	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	247	21.5	24.1	7.3	8.9	7.91	8.14	-	-	-
Confluence of Lindsey Sl. and Cache Sl. + 25 ppb PBO	181	21.5	24.1	7.3	8.5	7.71	8.07	-	-	-
San Joaquin River at Potato Slough + 25 ppb PBO	278	21.6	24.3	7.1	8.7	7.69	8.02	-	-	-
Old River, western arm at railroad bridge + 25 ppb PBO	595	21.6	24.1	7.4	8.7	7.79	8.04	-	-	-
Old River at mouth of Holland Cut + 25 ppb pBO	517	21.8	24.4	7.4	8.7	7.79	8.07	-	-	-

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 B 51-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 11/06/08 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 11/04/08 - 11/05/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	98	2.5	98	2.5	NS
Low EC Control @ 192.6 µS/cm	95	5.0	75	21.8	NS
High EC Control @ 14.01 mS/cm	100	0.0	80	20.0	NS
High EC Control @ 22.24 mS/cm[6]	95	2.9	70	20.0	NS
Napa River at River Park Blvd.[5]	97	3.1	89	6.4	NS
Sacramento River at Hood DWR Station[3]	100	0.0	91	9.4	NS
Rough and Ready DWR station, Stockton	100	0.0	93	7.5	NS
Suisun Slough at Rush Ranch[4]	100	0.0	90	7.1	NS
Suisun Bay off Chipps Island (508)[4]	95	2.8	63	15.8	NS
Montezuma Slough at Nurse Slough (609)[4]	100	0.0	85	15.0	NS
Grizzly Bay at Dolphin (602)[5]	98	2.5	89	11.1	NS
Field Dup.: Napa River at River Park Blvd.[5]	95	5.0	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.041	0.006	0.035	0.002	NS
Low EC Control @ 192.6 µS/cm	0.051	0.011	0.093	0.066	NS
High EC Control @ 14.01 mS/cm	0.039	0.002	0.027	0.005	NS
High EC Control @ 22.24 mS/cm[6]	0.028	0.003	0.037	0.023	NS
Napa River at River Park Blvd.[5]	0.026	0.006	0.022	0.009	NS
Sacramento River at Hood DWR Station[3]	0.091	0.006	0.035	0.010	S** (38%)
Rough and Ready DWR station, Stockton	0.091	0.008	0.041	0.011	S* (45%)
Suisun Slough at Rush Ranch[4]	0.067	0.011	0.046	0.009	NS
Suisun Bay off Chipps Island (508)[4]	0.052	0.002	0.039	0.007	NS
Montezuma Slough at Nurse Slough (609)[4]	0.067	0.006	0.065	0.011	NS
Grizzly Bay at Dolphin (602)[5]	0.047	0.003	0.034	0.010	NS
Field Dup.: Napa River at River Park Blvd.[5]	0.039	0.002	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

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

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

4. This high conductivity sample was compared to the High EC Control @ 14.01 mS/cm.

5. This high conductivity sample was compared to the High EC Control @ 22.24 mS/cm.

6. Two replicates in the PBO treatment were excluded from analysis because of high mortality at 48 hrs in a treatment with generally high survival. The treatment was not included in statistical analysis.

Table B 51-2. Summary of 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 11/04/08-11/05/08.

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 River Park Blvd.	20780	15.1	6.96	8.0	9.9	0.18	0.000
Sacramento River at Hood DWR Station	193	15.9	6.85	8.8	13.5	0.20	0.000
Rough and Ready DWR station, Stockton	680	17.3	7.34	8.2	6.6	0.15	0.001
Suisun Slough at Rush Ranch	13420	14.9	7.00	6.4	18.3	0.20	0.000
Suisun Bay off Chipps Island (508)	12180	15.8	7.84	9.7	7.8	0.12	0.002
Montezuma Slough at Nurse Slough (609)	12670	15.5	7.48	8.5	14.5	0.21	0.001
Grizzly Bay at Dolphin (602)	19130	15.8	7.80	10.1	10.7	0.11	0.001
Field Dup.: Napa River at River Park Blvd.	20780	15.1	6.96	8.0	5.4	0.19	0.000

Table B 51-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 11/06/08 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 11/04/08-11/05/08.

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	22.1	23.2	7.5	8.4	7.73	8.11	104	60	-
Low EC Control @ 192.6 µS/cm	223	22.3	23.2	7.5	8.5	7.48	7.95	64	32	-
High EC Control @ 14.01 mS/cm	13590	22.3	23.4	7.3	8.5	7.72	8.05	1680	48	-
High EC Control @ 22.24 mS/cm	21520	22.3	23.5	7.2	8.4	7.07	7.84	2680	90	-
Napa River at River Park Blvd.	19965	22.2	23.5	6.8	8.2	7.47	7.75	2560	92	0.002
Sacramento River at Hood DWR Station	220	22.3	22.7	7.2	8.6	7.66	7.90	76	76	0.004
Rough and Ready DWR station, Stockton	673	22.3	23.3	7.3	8.7	7.86	8.15	156	106	0.005
Suisun Slough at Rush Ranch	13020	22.2	23.3	7.2	8.1	7.41	8.24	1680	166	0.002
Suisun Bay off Chipps Island (508)	11665	22.0	23.5	7.2	8.3	7.77	7.96	1520	82	0.003
Montezuma Slough at Nurse Slough (609)	12205	22.3	23.5	7.2	8.4	7.57	7.99	1560	94	0.003
Grizzly Bay at Dolphin (602)	18495	22.3	23.8	7.0	8.4	7.75	7.99	2240	94	0.002
Field Dup.: Napa River at River Park Blvd.	19970	22.2	23.3	6.9	8.0	7.47	7.92	2480	90	0.002
DIEPAMHR	367	22.1	23.1	7.5	8.5	7.80	8.14	-	-	-
Low EC Control @ 192.6 µS/cm	217	22.4	23.4	7.5	8.5	7.55	8.02	-	-	-
High EC Control @ 14.01 mS/cm	6932	22.3	23.4	7.3	8.5	7.75	8.05	-	-	-
High EC Control @ 22.24 mS/cm	21300	22.3	23.3	7.3	8.0	7.25	8.05	-	-	-
Napa River at River Park Blvd.	20180	22.5	23.2	7.0	8.0	7.49	8.70	-	-	-
Sacramento River at Hood DWR Station	264	22.3	23.1	7.1	8.3	7.74	8.00	-	-	-
Rough and Ready DWR station, Stockton	690	23.1	23.2	7.1	8.1	7.89	8.15	-	-	-
Suisun Slough at Rush Ranch	12930	22.4	23.4	7.1	8.0	7.46	8.23	-	-	-
Suisun Bay off Chipps Island (508)	11450	22.3	23.4	7.1	8.3	7.76	7.99	-	-	-
Montezuma Slough at Nurse Slough (609)	12090	22.4	22.8	7.2	8.3	7.59	8.01	-	-	-
Grizzly Bay at Dolphin (602)	18420	22.9	23.5	7.0	8.1	7.78	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 B 52-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 11/07/2008 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 11/06/2008.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	90	4.1	97	2.8	NS
Low EC Control @ 201.3 uS/cm	98	2.5	98	2.5	NS
Sacramento R. Deep Water Channel, Light 55 [3]	98	2.5	98	2.5	NS
Sacramento River at tip of Grand Island (711) [3]	98	2.5	95	4.5	NS
Upper Cache Slough at mouth of Ulatis Creek	100	0.0	97	2.8	NS
Confluence of Lindsey Sl. and Cache Sl. [3]	95	3.1	83	14.4	NS
San Joaquin River at Potato Slough (815)	100	0.0	89	6.1	NS
Old River, western arm at railroad bridge (902)	100	0.0	98	2.5	NS
Old River at mouth of Holland Cut (915)	100	0.0	100	0.0	NS
Field Dup.: Upper Cache Slough at mouth of Ulatis Creek (557)	100	0.0	-	-	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.042	0.009	0.071	0.006	NS
Low EC Control @ 201.3 uS/cm	0.033	0.003	0.036*	0.006	NS
Sacramento R. Deep Water Channel, Light 55 [3]	0.069	0.006	0.086	0.013	NS
Sacramento River at tip of Grand Island (711) [3]	0.072	0.005	0.065	0.007	NS
Upper Cache Slough at mouth of Ulatis Creek	0.093	0.005	0.089	0.009	NS
Confluence of Lindsey Sl. and Cache Sl. [3]	0.060	0.011	0.066	0.010	NS
San Joaquin River at Potato Slough (815)	0.071	0.010	0.055	0.010	NS
Old River, western arm at railroad bridge (902)	0.086	0.003	0.067	0.012	NS
Old River at mouth of Holland Cut (915)	0.071	0.009	0.052	0.009	NS
Field Dup.: Upper Cache Slough at mouth of Ulatis Creek (557)	0.092	0.014	-	-	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

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

3. These low conductivity samples were compared to the Low EC Control @ 201.3 uS/cm

Table B 52-2. Summary of 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 11/06/2008.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (^oC)	pH	DO (mg/L)			
Sacramento R. Deep Water Channel, Light 55	212	15.1	7.76	9.6	14.3	0.21	0.003
Sacramento River at tip of Grand Island (711)	203	15.2	7.58	8.6	4.9	0.39	0.004
Upper Cache Slough at mouth of Ulatis Creek	449	14.6	7.84	9.0	23.3	0.14	0.002
Confluence of Lindsey Sl. And Cache Sl.	202	15.2	7.57	8.5	8.2	0.29	0.003
San Joaquin River at Potato Slough (815)	297	16.1	7.64	9.1	2.7	0.20	0.002
Old River, western arm at railroad bridge (902)	834	15.6	7.91	10.4	2.7	0.05	0.001
Old River at mouth of Holland Cut (915)	612	16.0	7.79	9.5	3.8	0.07	0.001
Field Dup.: Upper Cache Slough at mouth of Ulatis Creek (557)	449	14.6	7.84	9.0	23.8	0.10	0.002

Table B 52-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 11/07/2008 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 11/06/2008.

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 (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	341	22.9	24.1	7.4	8.5	7.75	8.00	104	60	-
Low EC Control @ 201.3 uS/cm	200	23.0	24.0	7.6	8.5	7.52	7.76	76	30	-
Sacramento R. Deep Water Channel, Light 55	203	23.0	24.0	7.3	8.7	7.68	7.99	76	74	0.007
Sacramento River at tip of Grand Island (711)	194	23.1	23.9	7.1	8.9	7.67	8.00	72	70	0.012
Upper Cache Slough at mouth of Ulatis Creek	437	23.1	24.0	7.3	8.4	7.96	8.19	136	118	0.007
Confluence of Lindsey Sl. And Cache Sl.	195	23.0	24.0	7.1	8.4	7.70	7.99	76	66	0.009
San Joaquin River at Potato Slough (815)	284	23.0	24.0	7.2	8.8	7.68	7.95	72	66	0.008
Old River, western arm at railroad bridge (902)	787	23.0	23.9	7.3	8.9	7.68	7.98	132	70	0.002
Old River at mouth of Holland Cut (915)	598	23.1	23.9	7.1	8.6	7.77	7.98	108	70	0.003
Field Dup.: Upper Cache Slough at mouth of Ulatis Creek (557)	442	23.0	24.0	7.0	8.3	7.89	8.20	148	118	0.004
DIEPAMHR + 25 ppb PBO	339	23.1	23.5	7.4	8.3	7.75	8.03	-	-	-
Low EC Control @ 201.3 uS/cm + 25 ppb PBO	200	23.0	23.6	7.4	8.5	7.53	7.85	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	205	23.0	23.9	7.1	8.4	7.70	7.99	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	196	23.0	23.8	7.1	8.8	7.66	8.00	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	437	23.3	23.7	7.4	8.4	7.95	8.21	-	-	-
Confluence of Lindsey Sl. And Cache Sl. + 25 ppb PBO	195	23.1	23.9	7.1	8.4	7.71	8.02	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb	288	23.8	23.8	7.2	8.5	7.67	7.98	-	-	-

PBO										
Old River, western arm at railroad bridge (902) + 25 ppb PBO	789	23.1	23.9	7.3	8.2	7.78	7.99	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	601	23.7	24.0	7.3	8.3	7.78	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 B 53-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 11/20/2008 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 11/18/2008 - 11/19/2008.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	100	0.0	100	0.0	NS
Low EC Control @ 205.3 uS/cm	95	2.9	97	2.8	NS
High EC Control @ 14.63 mS/cm	98	2.5	100	0.0	NS
High EC Control @ 21.88 mS/cm	95	5.0	90	7.1	NS
Suisun Slough at Rush Ranch[4]	100	0.0	100	0.0	NS
Sacramento River at Hood DWR Station[3]	100	0.0	98	2.5	NS
Rough and Ready DWR station, Stockton	100	0.0	100	0.0	NS
Suisun Bay off Chipps Island (508)[4]	100	0.0	100	0.0	NS
Montezuma Slough at Nurse Slough (609)[4]	98	2.5	100	0.0	NS
Grizzly Bay at Dolphin (602)[5]	98	2.5	100	0.0	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.060	0.006	0.051	0.004	NS
Low EC Control @ 205.3 uS/cm	0.050	0.007	0.049	0.004	NS
High EC Control @ 14.63 mS/cm	0.040*	0.005	0.036*	0.003	NS
High EC Control @ 21.88 mS/cm	0.030*	0.008	0.027*	0.005	NS
Suisun Slough at Rush Ranch[4]	0.083	0.004	0.070	0.004	NS
Sacramento River at Hood DWR Station[3]	0.068	0.005	0.072	0.003	NS
Rough and Ready DWR station, Stockton	0.091	0.005	0.090	0.009	NS
Suisun Bay off Chipps Island (508)[4]	0.057	0.010	0.057	0.003	NS
Montezuma Slough at Nurse Slough (609)[4]	0.049	0.002	0.051	0.005	NS
Grizzly Bay at Dolphin (602)[5]	0.040	0.003	0.037	0.006	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.
3. This low conductivity sample was compared to the Low EC Control.
4. This high conductivity sample was compared to the high EC Control @ 14.63 mS/cm.
5. This high conductivity sample was compared to the high EC Control @ 21.88 mS/cm.

Table B 53-2. Summary of 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 11/18/2008 - 11/19/2008.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (^{o}C)	pH	DO (mg/L)			
Suisun Slough at Rush Ranch	13130	13.7	7.33	7.4	33.6	0.22	0.001
Sacramento River at Hood DWR Station	204	15.6	7.26	9.4	9.1	0.45	0.002
Rough and Ready DWR station, Stockton	717	16.6	7.49	8.6	5.9	0.17	0.001
Suisun Bay off Chipps Island (508)	13000	15.1	7.64	9.7	35.6	0.15	0.001
Montezuma Slough at Nurse Slough (609)	14020	15.0	7.44	8.2	25.5	0.25	0.001
Grizzly Bay at Dolphin (602)	20920	15.3	7.76	9.4	30.8	0.15	0.002

Table B 53-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 11/20/2008 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 11/18/2008 - 11/19/2008.

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 (^{o}C)	Max Temp (^{o}C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	350	22.7	24.3	7.6	8.4	7.71	8.18	102	60	-
Low EC Control @ 205.3 uS/cm	207	22.8	24.1	7.7	8.4	7.46	8.11	64	46	-
High EC Control @ 14.63 mS/cm	14305	22.7	24.1	7.4	8.4	7.66	8.04	1680	82	-
High EC Control @ 21.88 mS/cm	20980	24.2	24.2	7.0	8.4	7.81	8.09	2560	104	-
Suisun Slough at Rush Ranch	12710	22.8	24.1	7.2	8.4	7.82	7.94	1540	181	0.005
Sacramento River at Hood DWR Station	208	22.7	24.1	7.3	8.5	7.75	8.06	74	81	0.019
Rough and Ready DWR station, Stockton	711	22.7	24.2	7.2	8.9	7.91	8.08	162	114	0.009

Suisun Bay off Chipps Island (508)	12885	22.6	24.2	7.4	8.4	7.72	7.88	1480	92	0.004
Montezuma Slough at Nurse Slough (609)	13735	22.7	24.1	7.1	8.1	7.74	7.84	1600	114	0.005
Grizzly Bay at Dolphin (602)	20515	22.7	24.2	7.1	7.9	7.66	7.86	2400	104	0.004
DIEPAMHR + 25 ppb PBO	356	22.6	24.0	7.2	8.3	7.77	8.19	-	-	-
Low EC Control @ 205.3 uS/cm + 25 ppb PBO	211	22.7	24.0	7.6	8.5	7.52	8.08	-	-	-
High EC Control @ 14.63 mS/cm + 25 ppb PBO	14300	22.6	24.0	7.4	8.3	7.67	8.04	-	-	-
High EC Control @ 21.88 mS/cm + 25 ppb PBO	20890	24.0	24.0	7.5	8.6	7.78	8.08	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	12645	22.8	23.9	7.1	8.4	7.74	7.95	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	214	22.8	23.9	7.4	8.4	7.75	8.16	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	714	22.9	23.9	7.3	8.7	7.97	8.01	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	12835	22.8	24.0	7.4	8.3	7.70	7.94	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	14200	24.0	24.4	7.3	8.3	7.74	7.85	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	20585	23.2	23.9	7.1	8.5	7.70	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 B 54-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 11/21/08 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 11/20/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	95	5.0	91	5.4	NS
Low E.C Control @ 221.8 µS/cm	98	2.5	100	0.0	NS
Sacramento River Deep Water Channel, Light 55 [3]	100	0.0	95	5.0	NS
Sacramento River at tip of Grand Island (711) [3]	100	0.0	89	6.1	NS
Upper Cache Slough at Mouth of Ulatis Creek	98	2.5	98	2.5	NS
Confluence of Lindsey Slough and Cache Slough [3]	100	0.0	88	8.9	NS
San Joaquin River at Potato Slough (815)	100	0.0	85	15.0	NS
Old River, Western Arm at Railroad Bridge (902)	94	5.6	100	0.0	NS
Old River at Mouth of Holland Cut (915)	93	7.5	100	0.0	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.047	0.013	0.035	0.005	NS
Low E.C Control @ 221.8 µS/cm	0.041	0.004	0.036	0.006	NS
Sacramento River Deep Water Channel, Light 55 [3]	0.066	0.009	0.055	0.008	NS
Sacramento River at tip of Grand Island (711) [3]	0.077	0.010	0.047	0.006	S* (61%)
Upper Cache Slough at Mouth of Ulatis Creek	0.071	0.006	0.063	0.002	NS

Confluence of Lindsey Slough and Cache Slough [3]	0.061	0.005	0.049	0.005	NS
San Joaquin River at Potato Slough (815)	0.065	0.004	0.070	0.016	NS
Old River, Western Arm at Railroad Bridge (902)	0.062	0.014	0.071	0.004	NS
Old River at Mouth of Holland Cut (915)	0.070	0.007	0.050	0.010	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

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

3. These low conductivity samples were compared to the Low EC Control.

Table B 54-2. Summary of 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 11/20/08.

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 Deep Water Channel, Light 55	246	15.0	7.32	9.6	29.8	0.18	0.001
Sacramento River at tip of Grand Island (711)	232	14.9	7.30	14.5	7.8	0.31	0.002
Upper Cache Slough at Mouth of Ulatis Creek	459	14.6	7.48	15.1	26.1	0.03	0.000
Confluence of Lindsey Slough and Cache Slough	224	15.0	7.36	14.4	12.5	0.29	0.002
San Joaquin River at Potato Slough (815)	401	15.3	7.33	13.8	3.8	0.14	0.001
Old River, Western Arm at Railroad Bridge (902)	865	15.2	7.33	14.9	3.9	0.05	0.000
Old River at Mouth of Holland Cut (915)	653	15.6	7.34	14.8	3.9	0.03	0.000

Table B 54-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 11/21/08 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 11/20/08.

Treatment	Laboratory Chemistry	Hardness	Alkalinity	Unionized

	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH	(mg/L as $CaCO_3$)	(mg/L as $CaCO_3$)	Ammonia (mg/L)[1]
DIEPAMHR	337	22.4	23.6	7.7	8.4	7.83	8.04	102	60	-
Low E.C Control @ 221.8 µS/cm	217	22.5	23.4	7.7	8.5	7.66	7.90	66	38	-
Sacramento River Deep Water Channel, Light 55	254	22.5	23.2	7.4	8.7	7.79	8.05	84	86	0.007
Sacramento River at tip of Grand Island (711)	231	22.5	23.3	7.6	8.7	7.77	8.06	82	82	0.012
Upper Cache Slough at Mouth of Ulatis Creek	451	22.5	22.8	7.5	8.6	8.02	8.22	140	126	0.001
Confluence of Lindsey Slough and Cache Slough	217	22.7	22.9	7.6	8.8	7.81	8.10	82	84	0.009
San Joaquin River at Potato Slough (815)	384	22.8	23.2	7.7	8.9	7.77	7.99	92	212	0.004
Old River, Western Arm at Railroad Bridge (902)	840	22.7	23.3	7.8	8.7	7.77	8.00	128	74	0.001
Old River at Mouth of Holland Cut (915)	634	22.5	23.3	7.7	8.6	7.83	7.97	114	76	0.001
DIEPAMHR + 25 ppb PBO	333	22.7	22.7	7.7	8.5	7.83	8.03	-	-	-
Low E.C Control @ 221.8 µS/cm + 25 ppb PBO	218	22.7	23.0	7.8	8.6	7.66	7.89	-	-	-
Sacramento River Deep Water Channel, Light 55 + 25 ppb PBO	255	22.8	23.1	7.7	8.7	7.79	8.05	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	230	22.9	23.1	7.4	8.6	7.83	8.05	-	-	-
Upper Cache Slough at Mouth of Ulatis Creek + 25 ppb PBO	460	23.1	23.2	7.6	8.8	8.04	8.24	-	-	-
Confluence of Lindsey Slough and Cache Slough + 25 ppb PBO	219	22.8	23.1	7.3	8.8	7.83	8.03	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	385	23.0	23.4	7.6	8.6	7.76	8.03	-	-	-
Old River, Western Arm at Railroad Bridge (902) + 25 ppb PBO	688	23.2	23.5	7.6	8.6	7.80	7.98	-	-	-
Old River at Mouth of Holland Cut (915) + 25 ppb PBO	633	23.2	23.2	7.8	8.8	7.85	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 B 55-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 12/04/08 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 12/02/08 - 12/03/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	90	5.8	100	0.0	NS
High E.C Control @ 14.16 mS/cm	98	2.5	100	0.0	NS
High E.C. Control @ 19.39 mS/cm	98	2.5	97	2.8	NS
Rough and Ready DWR station, Stockton	88	12.5	100	0.0	NS
Suisun Slough at Rush Ranch[3]	100	0.0	100	0.0	NS
Sacramento River at Hood DWR Station	95	5.0	95	3.1	NS
Montezuma Slough at Nurse Slough (609)[3]	100	0.0	100	0.0	NS
Grizzly Bay at Dolphin (602)[4]	100	0.0	100	0.0	NS
Suisun Bay off Chipps Island (508)[3]	98	2.5	100	0.0	NS

Field Dup.: Rough and Ready DWR station, Stockton	100	0.0	-	-	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.042	0.005	0.040	0.003	NS
High E.C Control @ 14.16 mS/cm	0.037	0.002	0.049	0.005	NS
High E.C. Control @ 19.39 mS/cm	0.036	0.002	0.032*	0.001	NS
Rough and Ready DWR station, Stockton	0.097	0.008	0.079	0.005	NS
Suisun Slough at Rush Ranch[3]	0.054	0.006	0.075	0.009	NS
Sacramento River at Hood DWR Station	0.074	0.005	0.064	0.009	NS
Montezuma Slough at Nurse Slough (609)[3]	0.072	0.008	0.049	0.006	NS
Grizzly Bay at Dolphin (602)[4]	0.044	0.003	0.038	0.004	NS
Suisun Bay off Chipps Island (508)[3]	0.052	0.005	0.048	0.005	NS
Field Dup.: Rough and Ready DWR station, Stockton	0.091	0.007	-	-	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

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

3. These high conductivity samples were compared to the High EC Control @ 14.16 mS/cm.

4. This high conductivity sample was compared to the High EC Control @ 19.39 mS/cm.

Table B 55-2. Summary of 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 12/02/08 - 12/03/08.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (^{o}C)	pH	DO (mg/L)			
Rough and Ready DWR station, Stockton	772	13.9	7.17	8.5	6.2	0.13	0.000
Suisun Slough at Rush Ranch	13920	11.5	7.00	7.2	41.7	0.23	0.000
Sacramento River at Hood DWR Station	222	12.7	7.12	9.6	5.1	0.54	0.002
Montezuma Slough at Nurse Slough (609)	12310	12.5	7.07	9.3	24.2	0.22	0.000
Grizzly Bay at Dolphin (602)	18570	13.1	7.57	10.0	10.4	0.15	0.001
Suisun Bay off Chipps Island (508)	11900	13.4	7.37	-	8.4	0.16	0.001
Field Dup.: Rough and Ready DWR station, Stockton	772	13.9	7.17	8.5	6.5	0.13	0.000

Table B 55-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 12/04/08 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 12/02/08 - 12/03/08.

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 (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	352	21.9	23.6	7.3	8.3	7.82	8.05	100	60	-
High E.C Control @ 14.16 mS/cm	13715	22.1	23.6	7.2	8.8	7.76	7.87	1600	74	-
High E.C. Control @ 19.39 mS/cm	18905	22.2	23.8	6.9	8.0	7.77	7.89	2200	80	-
Rough and Ready DWR station, Stockton	778	22.0	23.6	6.4	8.6	8.10	8.21	172	112	0.009
Suisun Slough at Rush Ranch	13540	22.2	23.9	6.9	8.9	7.68	8.21	1640	186	0.004
Sacramento River at Hood DWR Station	294	22.1	24.1	7.0	8.8	7.84	8.04	60	88	0.018
Montezuma Slough at Nurse Slough (609)	12350	22.1	24.0	7.1	9.0	7.76	7.98	1440	100	0.005
Grizzly Bay at Dolphin (602)	18400	22.0	23.9	7.0	8.7	7.75	7.92	2120	98	0.003
Suisun Bay off Chipps Island (508)	11730	22.1	23.8	7.4	8.3	7.81	7.92	1360	90	0.004
Field Dup.: Rough and Ready DWR station, Stockton	823	22.0	24.1	7.2	8.4	7.95	8.13	184	114	0.005
DIEPAMHR + 25 ppb PBO	381	22.1	23.6	7.5	8.4	7.82	8.12	-	-	-
High E.C Control @ 14.16 mS/cm + 25 ppb PBO	13630	22.1	23.9	7.3	8.7	7.77	7.86	-	-	-
High E.C. Control @ 19.39 mS/cm + 25 ppb PBO	19385	22.0	24.0	7.2	8.4	7.79	7.89	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	842	22.1	23.9	7.3	8.6	7.87	8.24	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	13355	22.0	23.9	7.2	8.8	7.69	8.21	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	288	22.1	23.8	6.9	8.4	7.88	8.04	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	12020	22.0	23.7	7.1	8.3	7.74	7.99	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	18250	22.0	23.9	7.0	8.6	7.73	7.88	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	11525	22.0	23.7	7.1	8.7	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 B 56-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 12/05/08 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 12/04/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	98	2.5	98	2.5	NS
Confluence of Lindsey Sl. and Cache Sl.	98	2.5	98	2.3	NS
Upper Cache Slough at mouth of Ulatis Creek	98	2.5	100	0.0	NS
Sacramento River at tip of Grand Island (711)	100	0.0	98	2.3	NS
San Joaquin River at Potato Slough (815)	98	2.5	100	0.0	NS
Old River at mouth of Holland Cut (915)	100	0.0	100	0.0	NS
Sacramento R. Deep Water Channel, Light 55	98	2.5	98	2.5	NS
Old River, western arm at railroad bridge (902)	98	2.3	100	0.0	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.041	0.008	0.035	0.006	NS
Confluence of Lindsey Sl. and Cache Sl.	0.065	0.005	0.047	0.005	S (72%)*
Upper Cache Slough at mouth of Ulatis Creek	0.091	0.012	0.064	0.011	NS
Sacramento River at tip of Grand Island (711)	0.082	0.011	0.080	0.006	NS
San Joaquin River at Potato Slough (815)	0.086	0.006	0.074	0.008	NS
Old River at mouth of Holland Cut (915)	0.083	0.010	0.086	0.009	NS
Sacramento R. Deep Water Channel, Light 55	0.062	0.003	0.068	0.003	NS
Old River, western arm at railroad bridge (902)	0.080	0.010	0.092	0.008	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

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

Table B 56-2. Summary of 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 12/04/08.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (^{o}C)	pH	DO (mg/L)			
Confluence of Lindsey Sl. and Cache Sl.	235	12.2	7.26	10.4	6.8	0.42	0.002
Upper Cache Slough at mouth of Ulatis Creek	432	11.5	7.52	11.3	18.7	0.07	0.000
Sacramento River at tip of Grand Island (711)	323	12.4	7.27	10.2	5.3	0.38	0.001
San Joaquin River at Potato Slough (815)	412	12.9	7.11	9.6	3.7	0.24	0.001
Old River at mouth of Holland Cut (915)	630	12.9	7.19	9.8	3.5	0.06	0.000
Sacramento R. Deep Water Channel, Light 55	251	12.2	7.30	10.4	8.2	0.33	0.001
Old River, western arm at railroad bridge (902)	818	12.6	7.18	10.4	3.1	0.07	0.000

Table B 56-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 12/05/08 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 12/04/08.

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 (^{o}C)	Max Temp (^{o}C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	339	21.6	22.7	7.6	8.4	7.70	8.05	100	60	-
Confluence of Lindsey Sl. and Cache Sl.	235	21.9	22.9	7.6	8.7	7.83	8.16	84	86	0.018
Upper Cache Slough at mouth of Ulatis Creek	431	22.0	23.0	7.2	8.6	8.01	8.24	140	122	0.003
Sacramento River at tip of Grand Island (711)	316	21.9	23.2	7.2	8.8	7.85	8.12	92	84	0.014
San Joaquin River at Potato Slough (815)	413	21.8	23.1	7.1	8.4	7.79	8.11	100	80	0.007
Old River at mouth of Holland Cut (915)	674	21.9	23.1	7.2	8.6	7.87	8.07	100	76	0.002
Sacramento R. Deep Water Channel, Light 55	250	21.9	23.3	7.2	8.8	7.87	8.13	84	88	0.015
Old River, western arm at railroad bridge (902)	825	22.1	23.4	7.5	8.6	7.81	8.03	156	76	0.002
DIEPAMHR + 25 ppb PBO	348	21.7	22.6	7.4	8.7	7.82	8.06	-	-	-
Confluence of Lindsey Sl. and Cache Sl. + 25 ppb PBO	234	21.7	23.0	7.2	8.6	7.86	8.14	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	432	21.7	22.9	7.4	8.9	8.06	8.25	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	320	21.7	23.1	7.0	8.9	7.85	8.12	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	420	21.8	23.0	7.3	8.9	7.86	8.09	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	652	21.5	23.0	7.6	8.8	7.89	8.09	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	239	21.9	23.1	7.1	8.9	7.85	8.14	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	798	21.8	23.3	7.7	8.8	7.87	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 B 57-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 12/18/08 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 12/16/08 - 12/17/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	100	0.0	100	0.0	NS
Low EC Control @ 123.6 µS/cm	98	2.5	92	5.3	NS
High EC Control @ 12.92 mS/cm	100	0.0	98	2.5	NS
High EC Control @ 20.46 mS/cm	98	2.5	95	2.9	NS
Suisun Slough at Rush Ranch	100	0.0	100	0.0	NS
Sacramento River at Hood DWR Station[3]	98	2.5	100	0.0	NS
Rough and Ready DWR station, Stockton	98	2.5	100	0.0	NS
Suisun Bay off Chipps Island (508)[4]	98	2.5	100	0.0	NS
Montezuma Slough at Nurse Slough (609)[4]	100	0.0	100	0.0	NS
Grizzly Bay at Dolphin (602)[5]	97	2.8	98	2.5	NS
Field Dup.: Montezuma Slough at Nurse Slough (609)[4]	100	0.0	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.029	0.005	0.038	0.004	NS
Low EC Control @ 123.6 µS/cm	0.026	0.001	0.032	0.003	NS
High EC Control @ 12.92 mS/cm	0.033	0.002	0.023*	0.005	NS
High EC Control @ 20.46 mS/cm	0.025	0.003	0.015**	0.004	NS
Suisun Slough at Rush Ranch	0.083	0.007	0.065	0.006	NS
Sacramento River at Hood DWR Station[3]	0.066	0.005	0.070	0.004	NS
Rough and Ready DWR station, Stockton	0.077	0.004	0.069	0.004	NS
Suisun Bay off Chipps Island (508)[4]	0.046	0.004	0.035	0.007	NS
Montezuma Slough at Nurse Slough (609)[4]	0.049	0.004	0.039	0.005	NS
Grizzly Bay at Dolphin (602)[5]	0.028	0.006	0.015	0.003	NS
Field Dup.: Montezuma Slough at Nurse Slough (609)[4]	0.059	0.003	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

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

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

4. This high conductivity sample was compared to the High EC Control @ 12.92 mS/cm.

5. This high conductivity sample was compared to the High EC Control @ 20.46 mS/cm.

Table B 57-2. Summary of 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 12/16/08-12/17/08.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Suisun Slough at Rush Ranch	13280	7.1	7.31	9.8	16.3	0.19	0.000
Sacramento River at Hood DWR Station	229	9.1	7.44	11.2	8.7	0.59	0.003
Rough and Ready DWR station, Stockton	960	10.5	7.66	10.1	9.2	0.17	0.001
Suisun Bay off Chipps Island (508)	12360	9.6	7.68	11.1	13.7	0.17	0.001
Montezuma Slough at Nurse Slough (609)	12360	8.4	7.54	10.8	23.3	0.19	0.001
Grizzly Bay at Dolphin (602)	19900	9.1	7.77	11.0	11.1	0.18	0.001
Field Dup.: Montezuma Slough at Nurse Slough (609)	12360	8.4	7.54	10.8	18.6	0.19	0.001

Table B 57-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 12/18/08 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 12/16/08-12/17/08.

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	21.8	7.4	8.5	7.85	8.18	96	60	-
Low EC Control @ 123.6 µS/cm	132	21.6	21.8	7.4	8.7	7.47	7.79	40	22	-
High EC Control @ 12.92 mS/cm	12585	21.8	22.5	7.3	8.2	7.79	7.92	1520	72	-
High EC Control @ 20.46 mS/cm	20325	21.8	22.6	7.6	8.0	7.77	7.89	2520	84	-
Suisun Slough at Rush Ranch	12870	21.7	22.8	7.4	8.5	7.81	8.16	1640	157	0.004
Sacramento River at Hood DWR Station	267	21.6	22.6	6.9	8.8	8.04	8.12	86	92	0.031
Rough and Ready DWR station, Stockton	966	21.6	23.0	7.3	8.6	8.14	8.25	202	113	0.012
Suisun Bay off Chipps Island (508)	12025	21.6	22.6	7.0	8.5	7.88	7.96	1560	94	0.004
Montezuma Slough at Nurse Slough (609)	11715	21.6	22.6	7.3	7.9	7.90	8.01	1600	94	0.006
Grizzly Bay at Dolphin (602)	19855	21.5	22.6	7.1	8.3	7.85	7.91	2560	98	0.004
Field Dup.: Montezuma Slough at Nurse Slough (609)	11365	21.5	23.0	7.3	8.3	7.86	7.98	1520	93	0.005
DIEPAMHR + 25 ppb PBO	1222	21.5	22.2	7.6	8.3	7.84	8.18	-	-	-
Low EC Control @ 123.6 µS/cm + 25 ppb PBO	133	21.5	22.1	7.4	8.4	7.45	7.94	-	-	-
High EC Control @ 12.92 mS/cm + 25 ppb PBO	12430	21.5	22.1	7.3	8.4	7.77	7.95	-	-	-
High EC Control @ 20.46 mS/cm + 25 ppb PBO	20285	21.5	22.2	7.1	8.0	7.78	7.89	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	13100	21.4	22.4	7.2	8.2	7.93	8.13	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	258	21.4	22.0	7.0	8.8	7.96	8.25	-	-	-

Rough and Ready DWR station, Stockton + 25 ppb PBO	957	21.4	22.8	7.3	8.7	8.15	8.20	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	11745	21.4	22.5	7.1	8.3	7.88	7.94	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	11645	21.4	22.9	7.3	8.2	7.86	7.95	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	19555	21.4	22.8	7.0	7.9	7.85	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 B 58-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 12/19/08 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 12/18/08.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	98	2.5	87	10.6	NS
Sacramento R. 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
Upper Cache Slough at mouth of Ulatis Creek	100	0.0	100	0.0	NS
Confluence of Lindsey Sl. and Cache Sl.	100	0.0	100	0.0	NS
San Joaquin River at Potato Slough (815)	100	0.0	100	0.0	NS
Old River, western arm at railroad bridge (902)	100	0.0	98	2.5	NS
Old River at mouth of Holland Cut (915)	100	0.0	100	0.0	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.037	0.003	0.053	0.004	NS
Sacramento R. Deep Water Channel, Light 55	0.086	0.003	0.073	0.010	NS
Sacramento River at tip of Grand Island (711)	0.061	0.006	0.073	0.001	NS
Upper Cache Slough at mouth of Ulatis Creek	0.074	0.007	0.077	0.008	NS
Confluence of Lindsey Sl. and Cache Sl.	0.075	0.005	0.076	0.008	NS
San Joaquin River at Potato Slough (815)	0.093	0.004	0.089	0.006	NS
Old River, western arm at railroad bridge (902)	0.093	0.003	0.096	0.005	NS
Old River at mouth of Holland Cut (915)	0.096	0.009	0.083	0.003	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

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

Table B 58-2. Summary of 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 12/18/08.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (^{o}C)	pH	DO (mg/L)			
Sacramento R. Deep Water Channel, Light 55	268	7.9	7.78	12.0	9.9	0.28	0.002
Sacramento River at tip of Grand Island (711)	259	8.1	7.65	11.5	6.2	0.51	0.003
Upper Cache Slough at mouth of Ulatis Creek	486	6.8	8.09	12.4	14.9	0.03	0.000
Confluence of Lindsey Sl. and Cache Sl.	266	7.6	7.78	12.1	8.3	0.34	0.003
San Joaquin River at Potato Slough (815)	436	8.7	7.60	11.5	5.0	0.24	0.001
Old River, western arm at railroad bridge (902)	953	7.4	7.83	12.3	4.4	0.05	0.000
Old River at mouth of Holland Cut (915)	737	7.9	7.80	12.3	4.1	0.03	0.000

Table B 58-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 12/19/08 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 12/18/08.

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 (^{o}C)	Max Temp (^{o}C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	331	22.0	22.6	7.7	8.7	7.82	8.06	96	60	-
Sacramento R. Deep Water Channel, Light 55	262	22.2	22.8	7.5	8.6	7.96	8.12	104	96	0.011
Sacramento River at tip of Grand Island (711)	249	22.4	23.2	7.6	8.6	8.00	8.11	88	94	0.023
Upper Cache Slough at mouth of Ulatis Creek	492	22.4	23.9	7.6	8.6	8.14	8.32	152	136	0.002
Confluence of Lindsey Sl. and Cache Sl.	254	22.4	23.7	7.6	8.6	7.96	8.11	96	98	0.014
San Joaquin River at Potato Slough (815)	420	22.4	23.8	7.7	8.7	7.91	8.07	108	88	0.009
Old River, western arm at railroad bridge (902)	929	22.3	24.6	7.8	8.8	7.89	8.07	152	88	0.002
Old River at mouth of Holland Cut (915)	723	22.3	24.4	7.7	8.6	7.82	8.10	132	84	0.001
DIEPAMHR + 25 ppb PBO	332	22.3	23.4	7.7	8.6	7.83	8.01	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	259	22.3	23.3	7.6	8.8	7.91	8.15	-	-	-

Sacramento River at tip of Grand Island (711) + 25 ppb PBO	248	22.3	23.7	7.6	8.7	7.87	8.09	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	485	22.2	23.5	7.7	8.8	8.06	8.32	-	-	-
Confluence of Lindsey Sl. and Cache Sl. + 25 ppb PBO	260	22.2	23.8	7.7	8.7	8.02	8.12	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	424	22.1	23.7	7.7	8.9	7.95	8.09	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	938	22.2	23.9	7.7	8.6	7.90	8.09	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	719	22.2	23.6	7.8	8.6	7.88	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 B 59-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 1/08/09 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 1/06/09 - 1/07/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	98	2.5	100	0.0	NS
High EC Control @ 12.46 mS/cm	98	2.5	100	0.0	NS
High EC Control @ 19.42 mS/cm	98	2.5	98	2.5	NS
Suisun Slough at Rush Ranch[3]	100	0.0	100	0.0	NS
Rough and Ready DWR station, Stockton	100	0.0	100	0.0	NS
Sacramento River at Hood DWR Station	98	2.5	100	0.0	NS
Napa River at River Park Blvd.[4]	100	0.0	100	0.0	NS
Suisun Bay off Chipps Island (508)[3]	100	0.0	100	0.0	NS
Grizzly Bay at Dolphin (602)[4]	98	2.5	93	4.8	NS
Field Dup.: Grizzly Bay at Dolphin (602)[4]	100	0.0	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.042	0.006	0.076	0.009	S (181%)*
High EC Control @ 12.46 mS/cm	0.037	0.005	0.061	0.007	S (165%)*
High EC Control @ 19.42 mS/cm	0.065	0.003	0.038	0.013	NS
Suisun Slough at Rush Ranch[3]	0.105	0.011	0.078	0.010	NS
Rough and Ready DWR station, Stockton	0.117	0.006	0.064	0.015	S (55%)*
Sacramento River at Hood DWR Station	0.100	0.004	0.088	0.005	NS
Napa River at River Park Blvd.[4]	0.032	0.007	0.041	0.005	NS
Suisun Bay off Chipps Island (508)[3]	0.066	0.006	0.106	0.008	S (161%)**
Grizzly Bay at Dolphin (602)[4]	0.028†	0.006	0.069	0.008	S (246%)**

Field Dup.: Grizzly Bay at Dolphin (602)[4]	0.047	0.014	-	-	NA	

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

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

3. These high conductivity samples were compared to the High EC Control @ 12.46 mS/cm.

4. These high conductivity samples were compared to the High EC Control @ 19.42 mS/cm.

†. This treatment showed lower weight compared to the High EC Control, but not compared to the normal EC Control.

Table B 59-2. Summary of 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 1/06/09 - 1/07/09.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Suisun Slough at Rush Ranch	11140	8.5	7.43	11.9	23.8	0.19	0.001
Rough and Ready DWR station, Stockton	983	8.7	7.32	11.2	5.6	0.10	0.000
Sacramento River at Hood DWR Station	216	8.2	7.25	11.3	13.0	0.46	0.002
Napa River at River Park Blvd.	18370	9.6	7.24	11.4	38.6	0.19	0.001
Suisun Bay off Chipps Island (508)	12330	8.3	7.37	10.2	13.1	0.26	0.001
Grizzly Bay at Dolphin (602)	19800	8.6	7.58	11.5	13.9	0.24	0.001
Field Dup.: Grizzly Bay at Dolphin (602)	19800	8.6	7.58	11.5	13.0	0.25	0.001

Table B 59-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 1/08/09 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 1/06/09 - 1/07/09.

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	355	21.0	23.4	7.2	8.7	7.80	7.97	104	62	-
High EC Control @ 12.46 mS/cm	11840	21.4	24.0	7.2	8.5	7.75	7.82	1380	74	-
High EC Control @ 19.42 mS/cm	17925	21.1	24.1	6.6	8.4	7.74	7.83	2200	83	-
Suisun Slough at Rush Ranch	10520	20.7	23.9	6.4	8.4	7.75	8.14	1320	164	0.004

Rough and Ready DWR station, Stockton	988	20.9	23.4	7.1	8.9	7.97	8.15	200	118	0.004
Sacramento River at Hood DWR Station	262	21.0	23.8	7.5	8.7	7.84	8.02	80	88	0.017
Napa River at River Park Blvd.	17400	20.9	24.1	7.6	8.4	7.62	7.91	2160	116	0.003
Suisun Bay off Chipps Island (508)	11545	20.8	23.4	7.6	8.5	7.74	7.93	1380	96	0.005
Grizzly Bay at Dolphin (602)	18230	20.5	23.7	6.7	8.4	7.72	7.86	2280	102	0.004
Field Dup.: Grizzly Bay at Dolphin (602)	18120	20.7	23.6	7.0	8.3	7.74	7.87	2180	102	0.005
DIEPAMHR + 25 ppb PBO	395	20.8	22.7	7.3	8.5	7.80	8.02	-	-	-
High EC Control @ 12.46 mS/cm + 25 ppb PBO	11485	20.5	22.7	7.3	8.5	7.76	7.82	-	-	-
High EC Control @ 19.42 mS/cm + 25 ppb PBO	18055	20.7	23.1	6.6	8.3	7.74	7.82	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	10140	20.4	22.6	7.5	8.4	7.70	8.14	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	978	20.6	22.8	7.7	8.7	7.98	8.15	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	262	20.5	22.9	7.5	8.7	7.85	8.02	-	-	-
Napa River at River Park Blvd. + 25 ppb PBO	17070	20.5	22.7	7.6	8.5	7.66	7.89	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	11135	20.2	22.4	7.0	8.6	7.76	7.92	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	18165	21.1	22.6	6.8	8.5	7.75	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 B 60-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 1/09/09 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 1/08/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	100	0.0	100	0.0	NS
Sacramento R. Deep Water Channel, Light 55	97	2.8	100	0.0	NS
Sacramento River at tip of Grand Island (711)	100	0.0	98	2.5	NS
Upper Cache Slough at mouth of Ulatis Creek	100	0.0	100	0.0	NS
Confluence of Lindsey Sl. And Cache Sl.	98	2.5	100	0.0	NS
San Joaquin River at Potato Slough (815)	100	0.0	100	0.0	NS
Old River, western arm at railroad bridge (902)	100	0.0	98	2.5	NS
Old River at mouth of Holland Cut (915)	100	0.0	100	0.0	NS
Trip Blank	100	0.0	-	-	NA

Treatment	Weight (mg/surviving individual)[1]

	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.069	0.002	0.056	0.008	NS
Sacramento R. Deep Water Channel, Light 55	0.105	0.008	0.106	0.008	NS
Sacramento River at tip of Grand Island (711)	0.085	0.012	0.097	0.014	NS
Upper Cache Slough at mouth of Ulatis Creek	0.124	0.008	0.107	0.004	NS
Confluence of Lindsey Sl. And Cache Sl.	0.111	0.010	0.096	0.010	NS
San Joaquin River at Potato Slough (815)	0.130	0.011	0.126	0.018	NS
Old River, western arm at railroad bridge (902)	0.129	0.007	0.129	0.006	NS
Old River at mouth of Holland Cut (915)	0.125	0.011	0.119	0.002	NS
Trip Blank	0.063	0.004	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

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

Table B 60-2. Summary of 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 1/08/09.

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	297	7.9	7.52	12.5	13.0	0.31	0.002
Sacramento River at tip of Grand Island (711)	313	7.9	7.42	12.1	11.2	0.39	0.002
Upper Cache Slough at mouth of Ulatis Creek	444	7.3	7.46	12.7	15.6	0.10	0.000
Confluence of Lindsey Sl. And Cache Sl.	261	7.9	7.34	12.1	11.0	0.39	0.001
San Joaquin River at Potato Slough (815)	474	7.8	7.24	12.0	4.7	0.25	0.001
Old River, western arm at railroad bridge (902)	784	7.6	7.48	11.8	4.5	0.12	0.001
Old River at mouth of Holland Cut (915)	745	7.6	7.47	12.6	3.8	0.12	0.000
Trip Blank	363	14.9	7.94	9.6	0.1	0.00	0.000

Table B 60-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 1/09/09 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 1/08/09.

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	329	21.2	22.6	7.0	8.3	7.76	8.04	104	62	-
Sacramento R. Deep Water Channel, Light 55	280	21.7	22.7	7.6	8.7	7.90	8.17	104	104	0.017
Sacramento River at tip of Grand Island (711)	258	21.7	23.1	7.3	8.7	7.83	8.18	84	86	0.012
Upper Cache Slough at mouth of Ulatis Creek	438	22.4	23.2	7.2	8.6	8.10	8.29	144	132	0.007
Confluence of Lindsey Sl. And Cache Sl.	245	21.1	22.7	7.4	8.5	7.85	8.11	94	97	0.021
San Joaquin River at Potato Slough (815)	433	22.5	22.8	6.9	8.6	7.79	8.15	100	84	0.013
Old River, western arm at railroad bridge (902)	715	21.9	22.5	7.1	8.6	7.91	8.06	136	88	0.005
Old River at mouth of Holland Cut (915)	691	22.2	22.6	7.5	8.4	7.90	8.11	132	85	0.006
Trip Blank	336	22.7	22.8	7.1	8.4	7.75	8.07	108	58	0.000
DIEPAMHR + 25 ppb PBO	340	22.7	22.7	7.6	8.2	7.76	8.08	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	289	22.4	23.0	7.5	8.3	7.90	8.15	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	278	22.7	22.9	7.5	8.5	7.80	8.11	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	448	22.7	22.9	6.8	8.8	8.05	8.27	-	-	-
Confluence of Lindsey Sl. And Cache Sl. + 25 ppb PBO	256	23.1	23.2	7.3	8.6	7.80	8.13	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	460	22.9	22.9	7.3	8.5	7.82	8.22	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	756	23.2	23.2	7.4	8.6	7.89	8.06	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	733	22.6	23.1	7.4	8.6	7.89	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 B 61-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 1/22/09 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 1/20/09 - 1/21/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	100	0.0	100	0.0	NS
High EC Control @ 12.68 mS/cm	100	0.0	98	2.5	NS
High EC Control @ 20.85 mS/cm	92	4.8	78*	4.8	NS
Suisun Slough at Rush Ranch[3]	100	0.0	100	0.0	NS

Rough and Ready DWR station, Stockton	98	2.5	100	0.0	NS
Napa River at River Park Blvd.[4]	100	0.0	100	0.0	NS
Suisun Bay off Chipps Island (508)[3]	100	0.0	100	0.0	NS
Grizzly Bay at Dolphin (602)[4]	100	0.0	97	2.8	NS
Montezuma Slough at Nurse Slough (609)	100	0.0	100	0.0	NS
Bottle Blank	100	0.0	-	-	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.075	0.008	0.067	0.009	NS
High EC Control @ 12.68 mS/cm	0.054*	0.003	0.040	0.005	S* (74%)
High EC Control @ 20.85 mS/cm	0.043**	0.003	0.057	0.004	S* (133%)
Suisun Slough at Rush Ranch[3]	0.118	0.003	0.121	0.008	NS
Rough and Ready DWR station, Stockton	0.087	0.011	0.115	0.007	NS
Napa River at River Park Blvd.[4]	0.068	0.009	0.080	0.003	NS
Suisun Bay off Chipps Island (508)[3]	0.045	0.006	0.073	0.004	S** (167%)
Grizzly Bay at Dolphin (602)[4]	0.040	0.005	0.060	0.001	S** (150%)
Montezuma Slough at Nurse Slough (609)	0.110	0.008	0.123	0.005	NS
Bottle Blank	0.062	0.005	-	-	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

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

3. These high conductivity samples were compared to the High EC Control @ 12.68 mS/cm.

4. These high conductivity samples were compared to the High EC Control @ 20.85 mS/cm.

Table B 61-2. Summary of 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 1/20/09 - 1/21/09.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Suisun Slough at Rush Ranch	11780	10.3	7.24	9.8	20.5	0.17	0.000
Rough and Ready DWR station, Stockton	1022	9.8	7.31	11.3	2.4	0.09	0.000

Napa River at River Park Blvd.	20870	11.9	7.46	10.4	36.7	0.11	0.000
Suisun Bay off Chipps Island (508)	12440	9.3	7.65	11.4	9.1	0.24	0.001
Grizzly Bay at Dolphin (602)	19140	9.6	7.69	11.7	8.3	0.22	0.001
Montezuma Slough at Nurse Slough (609)	7870	9.5	7.40	11.9	24.4	0.25	0.001
Bottle Blank	-	-	-	-	0.4	0.01	-

Table B 61-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 1/22/09 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 1/20/09 - 1/21/09.

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 (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	347	19.5	23.6	7.3	8.3	7.85	8.11	100	60	-
High EC Control @ 12.68 mS/cm	11945	19.3	23.5	7.1	8.8	7.74	7.98	1400	74	-
High EC Control @ 20.85 mS/cm	20050	20.3	23.5	7.2	8.3	7.73	7.98	2360	86	-
Suisun Slough at Rush Ranch	11210	20.4	23.2	7.2	8.6	7.60	8.23	1360	152	0.002
Rough and Ready DWR station, Stockton	1072	20.9	23.6	7.1	8.3	8.03	8.18	204	116	0.004
Napa River at River Park Blvd.	20080	21.0	23.6	6.8	8.5	7.63	8.00	2360	122	0.002
Suisun Bay off Chipps Island (508)	11900	20.0	23.5	6.9	8.5	7.72	8.02	1440	96	0.004
Grizzly Bay at Dolphin (602)	18730	21.5	23.7	7.0	8.3	7.81	7.94	2280	102	0.005
Montezuma Slough at Nurse Slough (609)	7660	21.0	23.7	6.9	8.5	7.77	8.02	880	96	0.005
Bottle Blank	367	20.8	23.9	7.1	8.9	7.83	8.09	108	58	0.001
DIEPAMHR + 25 ppb PBO	365	21.2	22.9	7.2	8.3	7.86	8.03	-	-	-
High EC Control @ 12.68 mS/cm + 25 ppb PBO	12215	21.9	22.9	7.1	8.2	7.74	7.95	-	-	-
High EC Control @ 20.85 mS/cm + 25 ppb PBO	20285	21.3	23.3	7.0	8.2	7.76	7.93	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	11330	21.0	23.3	6.8	8.3	7.66	8.15	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	1066	21.9	23.4	6.9	8.7	8.10	8.24	-	-	-
Napa River at River Park Blvd. + 25 ppb PBO	20315	21.6	23.3	6.8	8.0	7.59	7.97	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	11820	21.3	23.4	7.0	8.4	7.84	7.99	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	18775	22.0	23.3	7.1	8.0	7.71	7.95	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	7750	21.4	23.6	6.9	8.3	7.88	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 B 62-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 1/23/09 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 1/22/09 - 1/23/09.

Treatment	Survival (%)[1]	
	Unmanipulated	25 ppb PBO added

	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	92	4.8	95	3.1	NS
Low EC Control @ 136.5 uS/cm	98	2.3	97	2.8	NS
Confluence of Lindsey Sl. And Cache Sl.	98	2.5	100	0.0	NS
Old River at mouth of Holland Cut (915)	98	2.5	100	0.0	NS
San Joaquin River at Potato Slough (815)	98	2.5	100	0.0	NS
Upper Cache Slough at mouth of Ulatis Creek	98	2.5	100	0.0	NS
Sacramento R. Deep Water Channel, Light 55	95	2.9	100	0.0	NS
Old River, western arm at railroad bridge (902)	100	0.0	100	0.0	NS
Sacramento River at tip of Grand Island (711)	100	0.0	100	0.0	NS
Sacramento River at Hood DWR Station[3]	98	2.5	100	0.0	NS
Field Dup.: Sacramento River at tip of Grand Island (711)	98	2.5	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.075	0.006	0.053	0.009	NS
Low EC Control @ 136.5 uS/cm	0.112	0.005	0.067	0.007	S** (60%)
Confluence of Lindsey Sl. And Cache Sl.	0.115	0.018	0.105	0.013	NS
Old River at mouth of Holland Cut (915)	0.084	0.016	0.127	0.005	S* (151%)
San Joaquin River at Potato Slough (815)	0.122	0.006	0.126	0.006	NS
Upper Cache Slough at mouth of Ulatis Creek	0.125	0.012	0.135	0.012	NS
Sacramento R. Deep Water Channel, Light 55	0.115	0.005	0.083	0.013	NS
Old River, western arm at railroad bridge (902)	0.127	0.006	0.075	0.008	S** (59%)
Sacramento River at tip of Grand Island (711)	0.107	0.006	0.078	0.008	S* (73%)
Sacramento River at Hood DWR Station[3]	0.134	0.008	0.093	0.013	S* (69%)
Field Dup.: Sacramento River at tip of Grand Island (711)	0.098	0.009	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

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

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

Table B 62-2. Summary of 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 1/22/09 - 1/23/09.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (^{o}C)	pH	DO (mg/L)			
Confluence of Lindsey Sl. And Cache Sl.	271	10.2	7.61	11.3	6.4	0.35	0.003
Old River at mouth of Holland Cut (915)	741	9.5	7.51	11.5	4.0	0.04	0.000
San Joaquin River at Potato Slough (815)	557	9.5	7.42	11.4	5.1	0.18	0.001
Upper Cache Slough at mouth of Ulatis Creek	566	9.8	7.95	11.5	9.1	0.00	0.000
Sacramento R. Deep Water Channel, Light 55	303	10.0	7.90	11.2	14.1	0.25	0.003
Old River, western arm at railroad bridge (902)	830	9.4	7.52	11.4	4.2	0.05	0.000
Sacramento River at tip of Grand Island (711)	266	10.1	7.51	11.0	4.3	0.45	0.003
Sacramento River at Hood DWR Station	207	11.0	7.49	10.4	8.9	0.49	0.003
Field Dup.: Sacramento River at tip of Grand Island (711)	266	10.1	7.51	11.0	4.4	0.39	0.002

Table B 62-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 1/23/09 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 1/22/09 - 1/23/09.

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 (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	356	22.6	23.4	7.1	8.6	7.84	8.04	100	60	-
Low EC Control @ 136.5 uS/cm	154	22.5	23.8	7.3	8.8	7.45	7.87	44	24	-
Confluence of Lindsey Sl. And Cache Sl.	278	22.7	23.4	6.8	8.9	8.00	8.14	92	99	0.020
Old River at mouth of Holland Cut (915)	742	22.7	23.7	7.0	8.9	7.95	8.08	132	98	0.002
San Joaquin River at Potato Slough (815)	557	22.8	23.2	6.8	8.9	7.88	8.04	116	88	0.006
Upper Cache Slough at mouth of Ulatis Creek	569	22.8	23.0	7.1	8.9	8.07	8.34	168	150	0.000
Sacramento R. Deep Water Channel, Light 55	318	22.7	23.4	6.9	8.8	8.00	8.11	92	104	0.011
Old River, western arm at railroad bridge (902)	836	22.8	23.7	6.9	8.8	7.96	8.08	140	88	0.002
Sacramento River at tip of Grand Island (711)	258	22.8	23.7	6.7	8.9	7.84	8.00	84	88	0.014
Sacramento River at Hood DWR Station	217	22.8	23.5	6.9	8.7	7.60	8.01	72	78	0.009
Field Dup.: Sacramento River at tip of Grand Island (711)	278	22.8	23.6	6.8	8.6	7.90	8.05	84	81	0.020
DIEPAMHR + 25 ppb PBO	345	22.8	23.0	7.0	8.4	7.81	8.01	-	-	-
Low EC Control @ 136.5 uS/cm + 25 ppb PBO	156	22.8	23.5	7.1	8.7	7.49	7.86	-	-	-
Confluence of Lindsey Sl. And Cache Sl. + 25 ppb PBO	486	22.7	22.9	6.8	8.8	7.95	8.17	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	736	22.8	22.9	7.1	8.9	7.98	8.09	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	567	22.9	22.9	6.9	8.7	7.89	8.02	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	559	22.2	22.8	6.9	8.6	8.09	8.34	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	318	22.5	22.8	6.9	8.5	8.02	8.17	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	828	22.3	22.8	7.7	8.5	7.95	8.07	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	247	22.6	22.9	6.9	8.6	7.93	8.07	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	230	22.4	22.9	6.8	8.7	7.59	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 B 63-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 2/05/09 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 2/03/09 - 2/04/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	95	3.1	98	2.3	NS
High EC Control @ 10.21 mS/cm	100	0.0	100	0.0	NS
High EC Control @ 20.48 mS/cm	69**	6.8	83*	3.6	NS
Napa River at River Park Blvd.[4]	98	2.5	95	2.8	NS
Sacramento River at Hood DWR Station	95	2.9	97	2.8	NS
Suisun Slough at Rush Ranch[3]	100	0.0	100	0.0	NS
Rough and Ready DWR station, Stockton	100	0.0	98	2.5	NS
San Joaquin River at Potato Slough (815)	98	2.5	100	0.0	NS
Sacramento River at tip of Grand Island (711)	100	0.0	100	0.0	NS
Upper Cache Slough at mouth of Ulatis Creek	100	0.0	100	0.0	NS
Old River, western arm at railroad bridge (902)	95	2.9	98	2.5	NS
Confluence of Lindsey Sl. And Cache Sl.	100	0.0	98	2.5	NS
Old River at mouth of Holland Cut (915)	100	0.0	100	0.0	NS
Sacramento R. Deep Water Channel, Light 55	100	0.0	100	0.0	NS
Field Dup.: Sacramento R. Deep Water Channel, Light 55	100	0.0	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.066	0.009	0.052	0.003	NS
High EC Control @ 10.21 mS/cm	0.057	0.004	0.028*	0.008	S* (49%)
High EC Control @ 20.48 mS/cm	0.064	0.008	0.025***	0.002	S** (39%)
Napa River at River Park Blvd.[4]	0.066	0.007	0.022	0.012	S* (33%)
Sacramento River at Hood DWR Station	0.108	0.012	0.069	0.012	NS
Suisun Slough at Rush Ranch[3]	0.068	0.006	0.051	0.004	NS
Rough and Ready DWR station, Stockton	0.088	0.001	0.092	0.005	NS
San Joaquin River at Potato Slough (815)	0.117	0.012	0.087	0.006	NS
Sacramento River at tip of Grand Island (711)	0.104	0.005	0.087	0.014	NS
Upper Cache Slough at mouth of Ulatis Creek	0.121	0.011	0.063	0.005	S** (52%)
Old River, western arm at railroad bridge (902)	0.119	0.014	0.044	0.012	S** (37%)
Confluence of Lindsey Sl. And Cache Sl.	0.105	0.004	0.060	0.002	S*** (57%)
Old River at mouth of Holland Cut (915)	0.097	0.014	0.062	0.006	NS
Sacramento R. Deep Water Channel, Light 55	0.079	0.005	0.050	0.005	S** (63%)
Field Dup.: Sacramento R. Deep Water Channel, Light 55 †	0.051	0.005	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

***: $P < 0.001$

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 @ 20.48 mS/cm

†. The mean weight of animals exposed to the Field Duplicate of the Light 55 site was significantly lower than that of animals exposed to the original sample.

Table B 63-2. 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 2/03/09 - 2/04/09.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (^{o}C)	pH	DO (mg/L)			
Napa River at River Park Blvd.	20180	12.6	7.48	11.2	8.1	0.11	0.001
Sacramento River at Hood DWR Station	303	10.6	7.43	10.5	12.8	0.44	0.002
Suisun Slough at Rush Ranch	10090	11.5	7.45	11.1	24.0	0.10	0.000
Rough and Ready DWR station, Stockton	1045	11.0	7.89	11.3	3.7	0.05	0.001
San Joaquin River at Potato Slough (815)	572	10.5	7.56	11.4	4.0	0.16	0.001
Sacramento River at tip of Grand Island (711)	417	10.9	7.68	10.8	5.3	0.26	0.002
Upper Cache Slough at mouth of Ulatis Creek	490	10.7	8.40	13.8	9.1	0.00	0.000
Old River, western arm at railroad bridge (902)	723	10.9	7.60	11.1	3.8	0.06	0.000
Confluence of Lindsey Sl. And Cache Sl.	260	10.7	7.53	10.9	7.1	0.35	0.002
Old River at mouth of Holland Cut (915)	664	10.7	7.63	11.1	3.1	0.02	0.000
Sacramento R. Deep Water Channel, Light 55	278	10.6	7.57	11.5	8.2	0.33	0.002
Field Dup.: Sacramento R. Deep Water Channel, Light 55	278	10.6	7.57	11.5	7.6	0.38	0.003

Table 63-3. Water chemistry during a *H. azteca* initial screening toxicity test initiated on 2/05/09 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 2/03/09 - 2/04/09.

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	20.0	23.1	7.5	8.5	7.76	8.07	108	60	-
High EC Control @ 10.21 mS/cm	9570	19.7	23.5	7.6	8.6	7.80	8.08	1160	74	-
High EC Control @ 20.48 mS/cm	19625	20.5	23.3	7.4	8.1	7.79	8.04	2400	88	-
Napa River at River Park Blvd.	18730	20.9	23.3	7.0	8.1	7.72	8.19	2360	126	0.002
Sacramento River at Hood DWR Station	249	20.6	23.5	7.4	8.3	7.97	8.15	80	92	0.019
Suisun Slough at Rush Ranch	9445	20.4	23.4	7.3	8.3	7.77	8.37	1200	164	0.002
Rough and Ready DWR station, Stockton	967	21.1	23.2	7.4	8.3	8.09	8.32	212	116	0.003
San Joaquin River at Potato Slough (815)	543	20.1	23.3	7.6	8.4	7.80	8.26	124	94	0.004
Sacramento River at tip of Grand Island (711)	376	20.4	23.3	7.4	8.3	8.03	8.27	100	96	0.013
Upper Cache Slough at mouth of Ulatis Creek	474	20.8	23.3	7.5	8.5	8.23	8.55	148	146	0.000
Old River, western arm at railroad bridge (902)	707	21.2	23.4	7.7	8.6	7.90	8.21	140	92	0.002
Confluence of Lindsey Sl. And Cache Sl.	261	21.1	23.1	7.5	8.6	8.00	8.22	96	100	0.021
Old River at mouth of Holland Cut (915)	680	21.3	23.4	7.6	8.4	7.96	8.26	140	92	0.001
Sacramento R. Deep Water Channel, Light 55	275	21.1	23.4	7.5	8.6	8.07	8.90	104	112	0.021
Field Dup.: Sacramento R. Deep Water Channel, Light 55	268	21.2	23.4	7.2	8.3	8.02	8.29	96	104	0.018
DIEPAMHR + 25 ppb PBO	342	21.1	23.0	7.6	8.6	7.79	8.12	-	-	-
High EC Control @ 10.21 mS/cm + 25 ppb PBO	9685	21.4	23.2	7.6	8.3	7.80	8.05	-	-	-
High EC Control @ 20.48 mS/cm + 25 ppb PBO	19620	21.1	23.3	7.4	7.9	7.81	8.04	-	-	-
Napa River at River Park Blvd. + 25 ppb PBO	18885	21.3	23.4	7.2	8.1	7.81	8.19	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	236.6	21.1	23.3	7.4	8.5	7.94	8.19	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	9530	21.7	23.1	7.5	8.4	7.84	8.32	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	1011	20.9	23.1	7.6	8.6	8.08	8.36	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	549	21.5	23.0	7.6	8.8	7.98	8.22	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	376.85	21.3	23.5	7.4	8.8	7.96	8.22	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	487.65	21.6	23.1	7.5	8.4	8.23	8.54	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	716	21.4	22.8	7.6	8.6	7.95	8.22	-	-	-
Confluence of Lindsey Sl. And Cache Sl. + 25 ppb PBO	266.3	21.3	23.7	7.4	8.7	8.00	8.21	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	709	21.4	23.7	7.3	8.3	8.03	8.27	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	284.5	21.4	23.8	7.3	8.6	8.00	8.30	-	-	-

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 B 64-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 2/06/09 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 2/05/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	98	2.5	95	2.9	NS
High EC Control @ 13.57 mS/cm	98	2.5	97	2.8	NS
High EC Control @ 19.22 mS/cm	80	9.1	79	9.4	NS
Suisun Bay off Chipps Island (508)[3]	100	0.0	97	3.1	NS
Montezuma Slough at Nurse Slough (609)	100	0.0	98	2.5	NS
Grizzly Bay at Dolphin (602)[4]	94	3.4	98	2.5	NS
Trip Blank	98	2.5	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.046	0.004	0.027	0.006	S* (59%)
High EC Control @ 13.57 mS/cm	0.029*	0.006	0.034	0.004	NS
High EC Control @ 19.22 mS/cm	0.025**	0.005	0.035	0.011	NS
Suisun Bay off Chipps Island (508)[3]	0.028	0.004	0.046	0.003	S* (164%)
Montezuma Slough at Nurse Slough (609)	0.042	0.007	0.038	0.009	NS
Grizzly Bay at Dolphin (602)[4]	0.039	0.009	0.034	0.007	NS
Trip Blank	0.049	0.005	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

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

3. This high conductivity sample was compared to the High EC Control @ 13.57 mS/cm.

4. This high conductivity sample was compared to the High EC Control @ 19.22 mS/cm.

Table B 64-2. Summary of 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 2/05/09.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (^{o}C)	pH	DO (mg/L)			
Suisun Bay off Chipps Island (508)	12810	10.6	7.70	10.8	18.4	0.25	0.002
Montezuma Slough at Nurse Slough (609)	5140	10.7	7.55	10.8	29.0	0.27	0.001
Grizzly Bay at Dolphin (602)	17210	10.8	7.79	11.0	13.9	0.21	0.002
Trip Blank	345	16.1	8.01	10.1	0.3	0.00	0.000

Table B 64-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 2/06/09 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 2/05/09.

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 (^{o}C)	Max Temp (^{o}C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	327	22.1	24.1	7.7	8.2	7.83	8.09	108	60	-
High EC Control @ 13.57 mS/cm	13030	19.9	23.9	7.4	8.6	7.77	7.84	1760	80	-
High EC Control @ 19.22 mS/cm	18995	22.5	23.9	7.2	8.1	7.77	7.84	2280	86	-
Suisun Bay off Chipps Island (508)	13075	22.2	23.1	7.4	8.2	7.73	7.95	1800	106	0.005
Montezuma Slough at Nurse Slough (609)	5030	22.4	23.7	7.4	8.1	7.83	8.02	680	100	0.007
Grizzly Bay at Dolphin (602)	17090	21.8	23.0	7.0	8.1	7.71	7.94	2200	106	0.003
Trip Blank	355	22.5	23.9	7.6	8.6	7.80	8.15	108	48	0.000
DIEPAMHR + 25 ppb PBO	327	22.1	22.5	7.1	8.5	7.78	8.06	-	-	-
High EC Control @ 13.57 mS/cm + 25 ppb PBO	13090	22.1	23.3	7.4	8.2	7.77	7.82	-	-	-
High EC Control @ 19.22 mS/cm + 25 ppb PBO	18380	22.0	23.1	7.1	8.1	7.78	7.86	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	12365	20.9	23.6	7.2	8.5	7.77	7.96	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	5006	21.5	23.2	7.6	8.2	7.91	8.00	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	16875	20.4	23.0	7.2	8.2	7.74	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 B 65-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 2/19/09 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 2/17/09 - 2/18/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	95	2.9	97	2.8	NS
High EC Control @ 12.50 mS/cm	97	2.8	92	2.7	NS
High EC Control @ 21.92 mS/cm	69*	8.1	73	13.0	NS
High EC Control @ 24.63 mS/cm	54**	10.9	32*	10.5	NS
Napa River at River Park Blvd.	94	6.3	94	6.3	NS
Sacramento River at Hood DWR Station	95	2.9	93	4.8	NS
Suisun Slough at Rush Ranch	100	0.0	100	0.0	NS
Rough and Ready DWR station, Stockton	100	0.0	95	5.0	NS
Suisun Bay off Chipps Island (508)	100	0.0	100	0.0	NS
Montezuma Slough at Nurse Slough (609)	98	2.3	100	0.0	NS
Carquinez Strait, West of Benicia army dock (405)[4]	93	2.5	98	2.5	NS
Grizzly Bay at Dolphin (602)[3]	100	0.0	100	0.0	NS
Napa River at Vallejo Seawall (340)[5]	91	6.0	88	7.5	NS
Field Dup.: Rough and Ready DWR station, Stockton	100	0.0	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.039	0.004	0.032	0.006	NS
High EC Control @ 12.50 mS/cm	0.019**	0.003	0.026	0.005	NS
High EC Control @ 21.92 mS/cm	0.008***	0.002	0.009*	0.000	NS
High EC Control @ 24.63 mS/cm	0.025*	0.002	0.041	0.021	NS
Napa River at River Park Blvd.	0.052	0.002	0.044	0.004	NS
Sacramento River at Hood DWR Station	0.051	0.004	0.060	0.004	NS
Suisun Slough at Rush Ranch	0.035	0.006	0.060	0.001	S** (171%)
Rough and Ready DWR station, Stockton	0.040	0.003	0.050	0.004	NS
Suisun Bay off Chipps Island (508)	0.046	0.009	0.045	0.008	NS
Montezuma Slough at Nurse Slough (609)	0.036	0.004	0.043	0.003	NS
Carquinez Strait, West of Benicia army dock (405)[4]	0.020	0.003	0.033	0.003	NS
Grizzly Bay at Dolphin (602)[3]	0.031	0.003	0.023	0.003	NS

Napa River at Vallejo Seawall (340)[5]	0.023	0.004	0.052	0.007	S* (226%)
Field Dup.: Rough and Ready DWR station, Stockton	0.067	0.012	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

***: $P < 0.001$

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

Table B 65-2. Summary of 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 2/17/09 - 2/18/09.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (^{o}C)	pH	DO (mg/L)			
Napa River at River Park Blvd.	454	10.0	7.31	11.4	713.3	0.35	0.001
Sacramento River at Hood DWR Station	196	9.5	7.55	12.0	20.2	0.06	0.000
Suisun Slough at Rush Ranch	7310	9.9	7.47	10.4	41.4	0.08	0.000
Rough and Ready DWR station, Stockton	1107	11.0	7.93	11.2	3.7	0.43	0.006
Suisun Bay off Chipps Island (508)	6780	9.9	7.87	11.7	10.7	0.23	0.002
Montezuma Slough at Nurse Slough (609)	8000	9.8	7.65	11.4	25.0	0.21	0.001
Carquinez Strait, West of Benicia army dock (405)	23650	10.2	7.83	11.0	24.4	0.22	0.002
Grizzly Bay at Dolphin (602)	12200	10.0	7.85	11.6	13.1	0.23	0.002
Napa River at Vallejo Seawall (340)	22400	10.0	7.88	10.8	44.2	0.23	0.002
Field Dup.: Rough and Ready DWR station, Stockton	1107	11.0	7.93	11.2	3.5	0.06	0.001

Table B 65-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 2/19/09 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 2/17/09 - 2/18/09.

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	22.6	23.8	7.5	8.0	7.80	8.10	100	62	-
High EC Control @ 12.50 mS/cm	12110	22.7	24.0	7.3	8.0	7.76	7.90	1440	70	-
High EC Control @ 21.92 mS/cm	21710	22.5	24.1	7.1	7.7	7.38	7.82	2520	84	-
High EC Control @ 24.63 mS/cm	23825	22.7	23.9	6.8	7.6	7.69	7.91	2840	86	-
Napa River at River Park Blvd.	474	22.7	24.1	7.6	8.0	7.52	7.81	72	284	0.011
Sacramento River at Hood DWR Station	229	22.7	24.3	7.2	8.0	7.82	8.10	72	78	0.004
Suisun Slough at Rush Ranch	7045	22.7	23.8	7.3	8.0	7.16	8.17	840	172	0.003
Rough and Ready DWR station, Stockton	1102	22.7	23.5	7.3	8.4	7.16	8.18	212	240	0.027
Suisun Bay off Chipps Island (508)	6310	22.6	23.4	7.3	8.4	7.87	7.95	840	100	0.007
Montezuma Slough at Nurse Slough (609)	7595	22.6	23.8	7.3	8.2	7.84	7.99	880	92	0.008
Carquinez Strait, West of Benicia army dock (405)	20210	22.7	23.5	7.0	7.7	7.70	7.92	2400	104	0.006
Grizzly Bay at Dolphin (602)	11600	22.7	24.0	7.3	8.2	7.85	7.94	1480	102	0.007
Napa River at Vallejo Seawall (340)	22665	22.6	23.3	6.9	7.7	7.75	7.83	2760	104	0.005
Field Dup.: Rough and Ready DWR station, Stockton	1091	22.7	23.8	7.5	8.4	8.05	8.29	220	120	0.005
DIEPAMHR + 25 ppb PBO	1791	22.6	22.9	7.6	8.3	7.85	8.06	-	-	-
High EC Control @ 12.50 mS/cm + 25 ppb PBO	11925	22.6	23.3	7.4	8.0	7.74	7.92	-	-	-
High EC Control @ 21.92 mS/cm + 25 ppb PBO	21195	22.6	23.5	7.0	7.8	7.69	7.89	-	-	-
High EC Control @ 24.63 mS/cm + 25 ppb PBO	23640	22.6	23.3	6.9	7.7	7.68	7.91	-	-	-
Napa River at River Park Blvd. + 25 ppb PBO	450	22.6	23.2	7.5	8.2	7.56	7.87	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	209	22.7	23.3	7.1	8.2	7.84	8.19	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	6980	22.6	23.4	7.3	8.1	7.96	8.17	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	1079	22.5	23.5	7.3	8.3	8.08	8.20	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	6445	22.7	23.0	7.3	8.0	7.86	8.06	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	7605	22.6	23.2	7.3	8.1	7.84	7.90	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	19965	22.7	23.5	6.9	7.8	7.73	7.88	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	11825	22.6	23.2	7.3	8.2	7.84	7.95	-	-	-
Napa River at Vallejo Seawall (340) + 25 ppb PBO	23020	22.6	23.2	6.9	7.8	7.75	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 B 66-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 2/20/09 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 2/19/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	92	4.8	98	2.5	NS
Low EC Control @ 152.2 uS/cm	86	5.5	98	2.5	NS
Sacramento R. Deep Water Channel, Light 55	83	13.7	100	0.0	NS
Sacramento River at tip of Grand Island (711)[3]	95	3.1	70	23.4	NS
Old River, western arm at railroad bridge (902)	100	0.0	100	0.0	NS
San Joaquin River at Potato Slough (815)	95	2.9	100	0.0	NS
Upper Cache Slough at mouth of Ulatis Creek	98	2.5	95	3.1	NS
Old River at mouth of Holland Cut (915)	100	0.0	89	11.1	NS
Confluence of Linsey Sl. And Cache Sl.	100	0.0	95	2.9	NS
Bottle Blank	97	2.8	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.034	0.006	0.030	0.006	NS
Low EC Control @ 152.2 uS/cm	0.042	0.003	0.034	0.003	NS
Sacramento R. Deep Water Channel, Light 55	0.084	0.005	0.094	0.015	NS
Sacramento River at tip of Grand Island (711)[3]	0.055	0.012	0.068	0.006	NS
Old River, western arm at railroad bridge (902)	0.077	0.002	0.077	0.013	NS
San Joaquin River at Potato Slough (815)	0.056	0.005	0.098	0.007	S** (175%)
Upper Cache Slough at mouth of Ulatis Creek	0.074	0.008	0.042	0.006	S* (57%)
Old River at mouth of Holland Cut (915)	0.090	0.010	0.098	0.007	NS
Confluence of Linsey Sl. And Cache Sl.	0.085	0.010	0.143	0.060	NS
Bottle Blank	0.035	0.002	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

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

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

Table B 66-2. Summary of 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 2/19/09.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (^{o}C)	pH	DO (mg/L)			
Sacramento R. Deep Water Channel, Light 55	385	9.5	8.03	11.5	17.4	0.19	0.003
Sacramento River at tip of Grand Island (711)	145	8.9	7.38	11.0	82.8	0.33	0.001
Old River, western arm at railroad bridge (902)	590	10.5	7.88	11.3	5.1	0.09	0.001
San Joaquin River at Potato Slough (815)	354	10.5	7.74	11.0	5.6	0.25	0.002
Upper Cache Slough at mouth of Ulatis Creek	377	9.7	7.81	10.0	138.3	0.23	0.002
Old River at mouth of Holland Cut (915)	628	10.1	7.80	10.6	4.4	0.08	0.001
Confluence of Lindsey Sl. And Cache Sl.	300	9.3	7.82	11.0	30.8	0.23	0.002
Bottle Blank	-	-	-	-	0.2	0.00	-

Table B 66-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 2/20/09 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 2/19/09.

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 (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	326	21.1	23.9	7.6	8.0	7.78	8.04	100	62	-
Low EC Control @ 152.2 uS/cm	149	20.8	23.6	7.6	8.4	7.46	7.81	44	26	-
Sacramento R. Deep Water Channel, Light 55	365	22.2	23.4	7.3	8.2	8.05	8.28	120	104	0.015
Sacramento River at tip of Grand Island (711)	143	22.2	23.8	7.5	8.2	7.62	7.80	60	52	0.010
Old River, western arm at railroad bridge (902)	557	22.3	23.9	7.3	8.4	7.91	8.00	124	90	0.004
San Joaquin River at Potato Slough (815)	340	22.2	23.6	7.4	8.4	7.83	8.00	100	86	0.008
Upper Cache Slough at mouth of Ulatis Creek	369	22.2	23.7	7.3	8.3	7.92	8.12	124	100	0.012
Old River at mouth of Holland Cut (915)	604	22.0	23.5	7.5	8.2	7.92	8.05	128	90	0.003
Confluence of Lindsey Sl. And Cache Sl.	292	22.4	23.6	7.5	8.3	7.96	8.09	100	108	0.010
Bottle Blank	338	22.5	23.5	7.5	8.2	7.77	8.02	104	58	0.000
DIEPAMHR + 25 ppb PBO	330	22.4	22.4	7.4	8.2	7.78	8.01	-	-	-
Low EC Control @ 152.2 uS/cm + 25 ppb PBO	149	22.3	22.4	7.4	8.1	7.48	7.81	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	362	22.2	22.4	7.4	8.3	8.08	8.17	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	141	22.3	22.3	7.4	8.3	7.71	7.82	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	556	22.1	22.1	7.3	8.2	7.95	8.03	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	336	21.9	22.0	7.4	8.3	7.84	8.02	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	359	21.9	22.1	7.4	8.0	7.91	8.09	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	590	21.5	22.3	7.5	8.1	7.92	8.05	-	-	-
Confluence of Lindsey Sl. And Cache Sl. + 25 ppb PBO	289	21.7	22.1	7.4	8.4	7.95	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 B 67-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 3/05/09 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 3/03/09 - 3/04/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	95	2.9	98	2.5	NS
Napa River at River Park Blvd.	98	2.5	98	2.5	NS
Sacramento River at Hood DWR Station	100	0.0	98	2.3	NS
Suisun Slough at Rush Ranch	100	0.0	98	2.5	NS
Rough and Ready DWR station, Stockton	98	2.5	93	4.8	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
Carquinez Strait, West of Benicia army dock (405)	98	2.5	98	2.5	NS
Grizzly Bay at Dolphin (602)	100	0.0	100	0.0	NS
Napa River at Vallejo Seawall (340)	98	2.5	100	0.0	NS
Field Dup.: Napa River at Vallejo Seawall (340)	100	0.0	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.057	0.007	0.046	0.005	NS
Napa River at River Park Blvd.	0.128	0.005	0.102	0.013	NS
Sacramento River at Hood DWR Station	0.099	0.010	0.072	0.008	NS
Suisun Slough at Rush Ranch	0.100	0.004	0.087	0.013	NS
Rough and Ready DWR station, Stockton	0.126	0.008	0.106	0.008	NS
Suisun Bay off Chipps Island (508)	0.131	0.006	0.083	0.013	S* (63%)
Montezuma Slough at Nurse Slough (609)	0.123	0.005	0.093	0.014	NS
Carquinez Strait, West of Benicia army dock (405)	0.045	0.004	0.054	0.004	NS
Grizzly Bay at Dolphin (602)	0.100	0.007	0.101	0.006	NS
Napa River at Vallejo Seawall (340)	0.040	0.010	0.065	0.008	NS
Field Dup.: Napa River at Vallejo Seawall (340)	0.034*	0.009	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

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

Table B 67-2. Summary of 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 3/03/09 - 3/04/09

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 River Park Blvd.	237	12.0	7.33	10.7	88.9	0.09	0.000
Sacramento River at Hood DWR Station	187	12.8	7.28	9.5	43.9	0.25	0.001
Suisun Slough at Rush Ranch	2673	12.6	7.26	8.3	63.9	0.19	0.001
Rough and Ready DWR station, Stockton	878	13.2	7.42	8.7	5.8	0.15	0.001
Suisun Bay off Chipps Island (508)	401	12.1	7.35	10.4	39.8	0.16	0.001
Montezuma Slough at Nurse Slough (609)	2229	12.8	7.14	11.6	68.9	0.24	0.001
Carquinez Strait, West of Benicia army dock (405)	6510	11.9	7.47	13.0	115.3	0.24	0.001
Grizzly Bay at Dolphin (602)	1060	11.9	7.64	13.4	90.8	0.18	0.001
Napa River at Vallejo Seawall (340)	9460	11.9	7.52	13.0	77.9	0.23	0.001
Field Dup.: Napa River at Vallejo Seawall (340)	9460	11.9	7.52	13.0	76.2	0.23	0.001

Table B 67-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 3/05/09 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 3/03/09 - 3/04/09.

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	333	21.6	22.9	7.7	8.7	7.78	8.09	100	56	-
Napa River at River Park Blvd.	230	21.9	23.1	7.8	8.8	7.68	7.87	70	54	0.002
Sacramento River at Hood DWR Station	180	21.9	23.6	7.5	8.5	7.79	7.98	72	74	0.009
Suisun Slough at Rush Ranch	2556	21.9	23.9	7.6	8.7	7.70	8.14	380	130	0.004
Rough and Ready DWR station, Stockton	844	21.3	23.8	7.5	8.6	7.88	8.15	186	110	0.005
Suisun Bay off Chipps Island (508)	394	22.3	24.0	7.6	8.7	7.86	8.00	96	74	0.006
Montezuma Slough at Nurse Slough (609)	2159	21.6	23.7	7.5	8.5	7.05	7.94	292	80	0.006
Carquinez Strait, West of Benicia army dock (405)	6160	22.4	23.8	7.4	8.4	7.82	7.92	800	88	0.006
Grizzly Bay at Dolphin (602)	972	21.6	23.7	7.4	8.8	7.90	8.05	152	74	0.009
Napa River at Vallejo Seawall (340)	9110	22.1	23.9	7.6	8.9	7.78	7.90	1040	92	0.005
Field Dup.: Napa River at Vallejo Seawall (340)	8850	21.7	24.0	7.2	8.6	7.81	7.90	1160	94	0.006
DIEPAMHR + 25 ppb PBO	345	22.3	23.5	7.6	8.2	7.80	8.06	-	-	-
Napa River at River Park Blvd. + 25 ppb PBO	228	22.0	23.3	7.5	8.6	7.69	7.89	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	185	22.7	23.8	7.4	8.5	7.75	7.96	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	2608	22.0	23.6	7.5	8.5	7.88	8.15	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	847	21.9	24.0	7.4	8.6	7.95	8.14	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	397	22.5	23.7	7.6	8.9	7.83	7.97	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	2121	21.6	24.1	7.3	8.6	7.78	7.92	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	6175	22.4	24.1	7.4	8.4	7.82	7.94	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	966	21.9	24.0	7.6	8.6	7.87	8.00	-	-	-
Napa River at Vallejo Seawall (340) + 25 ppb PBO	9015	21.5	23.8	7.7	8.5	7.81	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 B 68-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 3/06/09 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 3/05/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	100	0.0	95	5.0	NS
Sacramento R. Deep Water Channel, Light 55	98	2.5	100	0.0	NS
Sacramento River at tip of Grand Island (711)	86	14.3	95	3.1	NS
Upper Cache Slough at mouth of Ulatis Creek	97	3.1	95	2.9	NS
Confluence of Linsey Sl. And Cache Sl.	98	2.5	95	2.9	NS
San Joaquin River at Potato Slough (815)	97	2.8	100	0.0	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	98	2.5	NS
Trip Blank	98	2.5	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.045	0.002	0.060	0.026	NS
Sacramento R. Deep Water Channel, Light 55	0.089	0.005	0.094	0.009	NS
Sacramento River at tip of Grand Island (711)	0.064	0.009	0.040	0.007	NS
Upper Cache Slough at mouth of Ulatis Creek	0.073	0.004	0.040	0.005	S** (55%)
Confluence of Linsey Sl. And Cache Sl.	0.083	0.009	0.068	0.004	NS
San Joaquin River at Potato Slough (815)	0.089	0.008	0.094	0.008	NS
Old River, western arm at railroad bridge (902)	0.101	0.007	0.086	0.009	NS
Old River at mouth of Holland Cut (915)	0.085	0.010	0.055	0.007	NS
Trip Blank	0.056	0.005	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

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

Table B 68-2. Summary of 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 3/05/09.

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	258	11.7	7.44	10.1	45.8	0.15	0.001
Sacramento River at tip of Grand Island (711)	137	11.4	7.10	10.1	146.3	0.20	0.000
Upper Cache Slough at mouth of Ulatis Creek	343	11.4	7.48	9.3	151.3	0.21	0.001
Confluence of Linsey Sl. And Cache Sl.	211	11.7	7.35	10.1	37.8	0.14	0.001
San Joaquin River at Potato Slough (815)	209	12.5	7.39	9.7	16.7	0.15	0.001
Old River, western arm at railroad bridge (902)	337	12.3	7.48	10.3	12.0	0.08	0.000
Old River at mouth of Holland Cut (915)	425	12.6	7.42	10.3	9.2	0.07	0.000
Trip Blank	335	16.9	8.03	9.0	0.3	0.00	0.000

Table B 68-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 3/06/09 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 3/05/09.

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	344	22.4	24.0	7.1	8.2	7.77	8.04	100	56	-
Sacramento R. Deep Water Channel, Light 55	261	22.4	23.9	7.3	8.3	7.83	7.94	106	90	0.005
Sacramento River at tip of Grand Island (711)	136	22.4	23.9	7.6	8.5	7.63	7.82	68	54	0.006
Upper Cache Slough at mouth of Ulatis Creek	347	22.4	23.9	7.3	8.2	7.96	8.08	128	112	0.009
Confluence of Linsey Sl. And Cache Sl.	215	22.2	23.9	7.3	8.3	7.83	7.94	78	78	0.005
San Joaquin River at Potato Slough (815)	216	22.1	24.0	7.2	8.5	7.73	7.83	84	60	0.004
Old River, western arm at railroad bridge (902)	344	22.2	24.3	7.6	8.2	7.87	7.94	84	88	0.003
Old River at mouth of Holland Cut (915)	423	22.2	24.0	7.5	8.5	7.86	7.95	108	84	0.003
Trip Blank	348	22.3	24.1	7.0	8.5	7.74	8.03	110	64	0.000
DIEPAMHR + 25 ppb PBO	343	22.2	22.9	7.4	8.1	7.73	8.03	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	259	22.2	22.9	7.2	8.6	7.83	7.99	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	135	22.2	22.8	7.4	8.4	7.66	7.86	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	339	22.2	22.7	7.4	8.5	8.00	8.14	-	-	-
Confluence of Linsey Sl. And Cache Sl. + 25 ppb PBO	212	22.2	22.6	7.6	8.8	7.83	7.98	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	210	22.2	22.4	7.5	8.6	7.78	7.85	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	340	22.2	22.4	7.0	8.4	7.86	7.92	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	412	22.0	22.4	7.4	8.5	7.88	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 B 69-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 3/19/09 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 3/17/09 - 3/18/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	95	2.8	98	2.5	NS
Suisun Bay off Chipps Island (508)	100	0.0	100	0.0	NS
Montezuma Slough at Nurse Slough (609)	100	0.0	91	6.4	NS
Grizzly Bay at Dolphin (602)	100	0.0	100	0.0	NS
Carquinez Strait, West of Benicia army dock (405)	98	2.5	100	0.0	NS
Napa River at Vallejo Seawall (340)	100	0.0	100	0.0	NS
Rough and Ready DWR station, Stockton	100	0.0	100	0.0	NS
Confluence of Lindsey Sl. and Cache Sl.	98	2.5	98	2.5	NS
Upper Cache Slough at mouth of Ulatis Creek	100	0.0	100	0.0	NS
Sacramento R. Deep Water Channel, Light 55	100	0.0	100	0.0	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.048	0.009	0.063	0.011	NS
Suisun Bay off Chipps Island (508)	0.098	0.006	0.074	0.009	NS
Montezuma Slough at Nurse Slough (609)	0.068	0.009	0.058	0.006	NS
Grizzly Bay at Dolphin (602)	0.077	0.004	0.071	0.007	NS
Carquinez Strait, West of Benicia army dock (405)	0.075	0.005	0.061	0.003	S* (81%)
Napa River at Vallejo Seawall (340)	0.073	0.002	0.057	0.005	S* (78%)
Rough and Ready DWR station, Stockton	0.093	0.006	0.064	0.007	S* (69%)
Confluence of Lindsey Sl. and Cache Sl.	0.062	0.009	0.065	0.004	NS
Upper Cache Slough at mouth of Ulatis Creek	0.069	0.002	0.077	0.008	NS
Sacramento R. Deep Water Channel, Light 55	0.072	0.004	0.097	0.003	S** (135%)

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

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

Table B 69-2. Summary of 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 3/17/09 - 3/18/09.

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)	358	13.6	6.91	10.3	38.4	0.13	0.000
Montezuma Slough at Nurse Slough (609)	2730	14.0	6.99	9.7	65.7	0.24	0.001
Grizzly Bay at Dolphin (602)	425	13.8	6.75	10.5	71.8	0.15	0.000
Carquinez Strait, West of Benicia army dock (405)	6750	13.2	7.14	10.5	97.9	0.20	0.001
Napa River at Vallejo Seawall (340)	11210	13.2	7.02	10.3	74.8	0.15	0.000
Rough and Ready DWR station, Stockton	740	15.7	7.68	8.5	6.0	0.08	0.001
Confluence of Lindsey Sl. and Cache Sl.	354	14.3	7.38	10.0	14.7	0.24	0.002
Upper Cache Slough at mouth of Ulatis Creek	614	14.3	7.97	9.9	32.1	0.08	0.002
Sacramento R. Deep Water Channel, Light 55	369	13.0	7.79	10.0	18.7	0.18	0.003

Table B 69-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 3/19/09 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 3/17/09 - 3/18/09.

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 (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	332	21.8	23.5	7.6	8.4	7.77	8.10	100	58	-
Suisun Bay off Chipps Island (508)	370	21.7	23.7	7.6	8.4	7.81	8.04	92	72	0.006
Montezuma Slough at Nurse Slough (609)	2587	21.6	23.9	7.5	8.4	7.77	7.86	380	78	0.007
Grizzly Bay at Dolphin (602)	420	21.6	23.8	7.6	8.5	7.86	8.06	320	72	0.008
Carquinez Strait, West of Benicia army dock (405)	6590	21.3	24.0	7.5	8.6	7.77	7.88	800	80	0.005
Napa River at Vallejo Seawall (340)	10390	21.6	24.3	7.3	8.7	7.76	7.91	1260	88	0.005
Rough and Ready DWR station, Stockton	723	21.5	23.6	7.2	8.4	7.98	8.14	176	104	0.004
Confluence of Lindsey Sl. and Cache Sl.	283	21.5	24.2	7.5	8.8	7.89	8.18	100	100	0.016
Upper Cache Slough at mouth of Ulatis Creek	577	21.4	23.8	7.6	8.7	8.14	8.38	212	180	0.005
Sacramento R. Deep Water Channel, Light 55	355	21.5	23.6	7.5	8.4	8.04	8.21	124	124	0.013
DIEPAMHR + 25 ppb PBO	332	21.5	23.0	7.8	8.1	7.80	8.09	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	352	21.9	23.2	7.5	8.6	7.81	8.02	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	2530	21.7	23.1	7.5	8.3	7.70	7.84	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	418	21.9	23.2	7.6	8.5	7.84	8.08	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	6410	21.7	23.0	6.3	8.2	7.54	7.91	-	-	-
Napa River at Vallejo Seawall (340) + 25 ppb PBO	10275	21.7	23.4	7.3	8.2	7.73	7.83	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	710	21.5	23.4	7.7	8.5	7.94	8.15	-	-	-
Confluence of Lindsey Sl. and Cache Sl. + 25 ppb PBO	286	21.4	23.4	7.6	8.7	7.90	8.23	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	575	21.2	23.5	7.6	8.9	8.27	8.37	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	357	21.5	23.6	7.4	8.6	8.00	8.26	-	-	-

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 B 70-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 3/20/09 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 3/18/09 - 3/19/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	92	2.6	94	5.6	NS
Sacramento River at tip of Grand Island (711)	95	2.9	95	2.9	NS
San Joaquin River at Potato Slough (815)	100	0.0	100	0.0	NS
Old River, western arm at railroad bridge (902)	100	0.0	98	2.5	NS
Old River at mouth of Holland Cut (915)	100	0.0	98	2.5	NS
Napa River at River Park Blvd.	100	0.0	100	0.0	NS
Suisun Slough at Rush Ranch	100	0.0	100	0.0	NS
Sacramento River at Hood DWR Station	95	5.0	95	2.9	NS
Instant Ocean Control @ 150 mS/cm	98	2.5	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.026	0.005	0.046	0.005	S* (177%)
Sacramento River at tip of Grand Island (711)	0.061	0.005	0.061	0.008	NS
San Joaquin River at Potato Slough (815)	0.046	0.004	0.087	0.007	S** (189%)
Old River, western arm at railroad bridge (902)	0.064	0.013	0.065	0.010	NS
Old River at mouth of Holland Cut (915)	0.093	0.006	0.069	0.004	S* (74%)
Napa River at River Park Blvd.	0.084	0.011	0.100	0.012	NS
Suisun Slough at Rush Ranch	0.074	0.005	0.093	0.006	S* (126%)
Sacramento River at Hood DWR Station	0.092	0.003	0.067	0.006	S* (73%)
Instant Ocean Control @ 150 mS/cm	0.033	0.004	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

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

Table B 70-2. Summary of 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 3/18/09 - 3/19/09.

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)	210	14.1	7.16	10.1	5.9	0.34	0.001
San Joaquin River at Potato Slough (815)	218	14.4	6.94	10.0	10.3	0.11	0.000
Old River, western arm at railroad bridge (902)	258	14.8	7.39	10.1	10.9	0.05	0.000
Old River at mouth of Holland Cut (915)	319	14.8	7.21	9.8	7.6	0.03	0.000
Napa River at River Park Blvd.	466	16.6	7.96	10.5	42.3	0.05	0.001
Suisun Slough at Rush Ranch	4106	19.0	7.42	9.2	98.5	0.16	0.001
Sacramento River at Hood DWR Station	197	14.4	7.14	9.9	5.4	0.46	0.002

Table B 70-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 3/20/09 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 3/18/09 - 3/19/09.

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 (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	334	22.1	23.9	7.6	8.4	7.29	8.14	100	58	-
Sacramento River at tip of Grand Island (711)	199	22.1	23.8	7.4	8.4	7.29	8.08	80	78	0.015
San Joaquin River at Potato Slough (815)	206	22.3	24.0	7.3	8.7	7.20	8.17	84	68	0.004
Old River, western arm at railroad bridge (902)	254	22.3	23.6	7.5	8.8	7.27	8.14	84	68	0.002
Old River at mouth of Holland Cut (915)	286	22.3	23.8	7.3	8.5	7.34	8.11	88	70	0.000
Napa River at River Park Blvd.	453	22.4	23.8	7.3	8.9	7.51	8.20	88	96	0.003
Suisun Slough at Rush Ranch	3885	22.3	23.9	6.9	8.4	7.85	8.31	620	222	0.009
Sacramento River at Hood DWR Station	219	22.4	24.0	6.9	8.7	7.28	8.22	72	80	0.029
Instant Ocean Control @ 150 mS/cm	156	22.3	23.5	7.5	8.3	5.94	8.69	40	5	-
DIEPAMHR + 25 ppb PBO	338	22.2	23.3	7.4	8.5	7.33	8.26	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	204	22.3	23.5	7.6	8.5	7.39	8.25	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	207	22.4	23.6	7.4	8.7	7.18	8.15	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	244	22.5	23.7	7.3	8.7	7.29	8.10	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	289	22.3	23.4	7.2	8.6	7.34	8.14	-	-	-
Napa River at River Park Blvd. + 25 ppb PBO	450	22.3	23.7	7.3	8.6	7.51	8.23	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	4056	22.4	23.4	7.2	8.3	7.90	8.31	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	200	22.5	23.6	6.9	8.5	7.31	8.24	-	-	-

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 B 71-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 4/02/09 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 3/31/09 - 4/01/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	95	5.0	95	3.1	NS
High EC Control @ 19.70 mS/cm	89	4.2	89	7.0	NS
Sacramento River at Hood DWR Station	100	0.0	94	5.6	NS
Napa River at River Park Blvd.	98	2.5	98	2.5	NS
Suisun Slough at Rush Ranch	100	0.0	100	0.0	NS
Rough and Ready DWR station, Stockton	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)	95	2.9	100	0.0	NS
Suisun Bay off Chipps Island (508)	98	2.1	100	0.0	NS
Carquinez Strait, West of Benicia army dock (405)	94	3.7	94	3.4	NS
Napa River at Vallejo Seawall (340)	100	0.0	95	3.1	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.032	0.003	0.046	0.007	NS
High EC Control @ 19.70 mS/cm	0.032	0.005	0.039	0.009	NS
Sacramento River at Hood DWR Station	0.064	0.005	0.079	0.003	NS
Napa River at River Park Blvd.	0.084	0.005	0.073	0.005	NS
Suisun Slough at Rush Ranch	0.074	0.008	0.091	0.007	NS
Rough and Ready DWR station, Stockton	0.094	0.008	0.102	0.010	NS
Grizzly Bay at Dolphin (602)	0.094	0.006	0.085	0.010	NS
Montezuma Slough at Nurse Slough (609)	0.088	0.003	0.091	0.002	NS
Suisun Bay off Chipps Island (508)	0.087	0.008	0.130	0.017	NS
Carquinez Strait, West of Benicia army dock (405)	0.046	0.007	0.056	0.007	NS
Napa River at Vallejo Seawall (340)	0.059	0.009	0.053	0.003	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

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

Table B 71-2. Summary of 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 3/31/09 - 4/01/09.

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 Hood DWR Station	178	14.7	6.91	9.9	6.9	0.43	0.001
Napa River at River Park Blvd.	1430	17.4	7.77	9.6	105.0	0.22	0.004
Suisun Slough at Rush Ranch	3805	16.6	7.45	9.0	343.0	0.27	0.002
Rough and Ready DWR station, Stockton	913	16.3	7.94	10.1	7.2	0.02	0.000
Grizzly Bay at Dolphin (602)	5260	16.9	7.67	10.0	68.4	0.10	0.001
Montezuma Slough at Nurse Slough (609)	3439	15.9	7.12	9.0	80.3	0.25	0.001
Suisun Bay off Chipps Island (508)	5300	14.8	7.55	10.3	40.4	0.16	0.001
Carquinez Strait, West of Benicia army dock (405)	17740	15.7	7.53	9.8	155.7	0.21	0.001
Napa River at Vallejo Seawall (340)	18760	15.6	6.94	9.6	37.5	0.12	0.000

Table B 71-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 4/02/09 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 3/01/09 - 4/01/09.

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 (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	334	22.8	22.8	7.4	8.2	7.77	8.06	100	56	-
High EC Control @ 19.70 mS/cm	19135	22.4	23.1	7.0	8.2	7.72	7.78	2100	82	-
Sacramento River at Hood DWR Station	218	22.8	23.1	7.0	8.5	7.63	8.13	64	72	0.012
Napa River at River Park Blvd.	1325	22.1	23.1	7.3	8.5	7.89	8.12	240	118	0.007
Suisun Slough at Rush Ranch	3705	22.5	23.1	7.4	7.9	7.85	8.27	496	176	0.007
Rough and Ready DWR station, Stockton	931	23.1	23.1	7.3	8.3	7.95	8.14	164	104	0.001
Grizzly Bay at Dolphin (602)	4760	23.1	23.1	7.4	8.1	7.83	7.96	500	86	0.003
Montezuma Slough at Nurse Slough (609)	3382	22.7	23.1	7.4	8.4	7.71	8.00	388	84	0.005
Suisun Bay off Chipps Island (508)	5255	23.1	23.7	7.5	7.9	7.82	7.99	564	86	0.004
Carquinez Strait, West of Benicia army dock (405)	16675	23.1	23.4	7.1	8.1	7.65	7.88	1860	98	0.004
Napa River at Vallejo Seawall (340)	18515	23.1	23.9	7.2	7.7	7.65	7.92	1996	94	0.002
DIEPAMHR + 25 ppb PBO	391	22.6	23.0	7.3	7.9	7.77	8.03	-	-	-
High EC Control @ 19.70 mS/cm + 25 ppb PBO	19055	22.9	23.2	6.9	7.8	7.73	7.80	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	240	23.1	23.8	7.2	8.1	7.70	7.97	-	-	-
Napa River at River Park Blvd. + 25 ppb PBO	1362	23.2	23.4	7.4	8.2	7.93	8.15	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	3699	23.1	23.9	7.2	8.2	7.89	8.27	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	916	23.1	23.5	7.3	8.5	7.96	8.17	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	4656	23.1	24.1	7.3	8.1	7.81	7.95	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	3458	23.5	23.5	7.0	8.5	7.76	7.98	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	5165	23.3	24.0	7.1	8.3	7.81	7.96	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	16845	23.2	23.6	6.9	8.3	7.66	7.87	-	-	-
Napa River at Vallejo Seawall (340) + 25 ppb PBO	18575	23.2	23.9	7.0	8.1	7.70	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 B 72-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 4/03/09 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 4/02/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	100	0.0	76	17.9	NS
Sacramento R. Deep Water Channel, Light 55	98	2.5	100	0.0	NS
Upper Cache Slough at mouth of Ulatis Creek	100	0.0	98	2.5	NS
Confluence of Linsey Sl. And Cache Sl.	98	2.5	98	2.5	NS
Sacramento River at tip of Grand Island (711)	95	2.9	98	2.5	NS
San Joaquin River at Potato Slough (815)	98	2.5	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)	98	2.5	98	2.5	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.050	0.008	0.050	0.012	NS
Sacramento R. Deep Water Channel, Light 55	0.068	0.022	0.112	0.005	NS
Upper Cache Slough at mouth of Ulatis Creek	0.036	0.005	0.106	0.005	S*** (294%)
Confluence of Linsey Sl. And Cache Sl.	0.083	0.006	0.088	0.005	NS
Sacramento River at tip of Grand Island (711)	0.076	0.002	0.088	0.007	NS
San Joaquin River at Potato Slough (815)	0.090	0.003	0.105	0.006	NS
Old River, western arm at railroad bridge (902)	0.090	0.007	0.124	0.007	S* (138%)
Old River at mouth of Holland Cut (915)	0.100	0.013	0.105	0.006	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

***: $P < 0.001$

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

Table B 72-2. Summary of 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 4/02/09.

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	283	14.9	7.02	10.1	19.5	0.26	0.001
Upper Cache Slough at mouth of Ulatis Creek	605	15.3	7.96	10.2	29.3	0.04	0.001
Confluence of Linsey Sl. And Cache Sl.	272	15.5	6.90	9.9	14.2	0.28	0.001
Sacramento River at tip of Grand Island (711)	217	16.3	7.38	9.7	8.5	0.38	0.003
San Joaquin River at Potato Slough (815)	232	16.0	7.50	10.0	9.6	0.10	0.001
Old River, western arm at railroad bridge (902)	262	15.9	7.49	9.7	12.3	0.04	0.000
Old River at mouth of Holland Cut (915)	268	15.9	7.41	9.8	9.0	0.01	0.000

Table B 72-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 4/03/09 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 4/02/09.

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	22.1	23.5	7.5	8.5	7.69	8.12	100	56	-
Sacramento R. Deep Water Channel, Light 55	264	22.3	23.2	7.3	8.9	7.78	8.10	104	92	0.012
Upper Cache Slough at mouth of Ulatis Creek	571	22.3	23.5	7.3	8.6	8.24	8.39	196	172	0.003
Confluence of Linsey Sl. And Cache Sl.	248	22.6	23.4	7.0	8.8	7.79	8.13	92	88	0.017
Sacramento River at tip of Grand Island (711)	196	22.7	23.5	7.3	8.8	7.72	8.04	80	74	0.019
San Joaquin River at Potato Slough (815)	222	22.6	23.5	7.3	8.3	7.72	8.05	100	72	0.005
Old River, western arm at railroad bridge (902)	253	22.7	23.4	7.5	8.8	7.79	8.06	84	72	0.002
Old River at mouth of Holland Cut (915)	256	22.7	23.3	7.4	8.8	7.75	8.07	88	74	0.001
DIEPAMHR + 25 ppb PBO	334	22.7	23.5	7.6	8.3	7.72	8.05	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	258	22.6	23.1	7.0	8.4	7.74	8.06	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	567	22.7	23.3	7.2	8.5	8.23	8.37	-	-	-
Confluence of Linsey Sl. And Cache Sl. + 25 ppb PBO	245	22.6	22.8	7.0	8.5	7.79	8.05	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	197	22.7	23.1	7.3	8.5	7.19	8.07	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	232	22.6	23.2	7.3	8.4	7.74	8.05	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	253	22.6	22.9	7.4	9.0	7.81	8.12	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb	258	22.7	23.1	7.2	8.8	7.78	7.97	-	-	-

PBO

Table B 73-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 4/16/09 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 4/14/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	98	2.5	83	11.1	NS
High EC Control @ 19.88 mS/cm	87	6.3	81	10.8	NS
Rough and Ready DWR station, Stockton	100	0.0	97	2.8	NS
Montezuma Slough at Nurse Slough (609)	100	0.0	95	2.9	NS
Grizzly Bay at Dolphin (602)	95	2.7	100	0.0	NS
Napa River at Vallejo Seawall (340)[3]	95	3.1	88	4.8	NS
Carquinez Strait, West of Benicia army dock (405)	100	0.0	98	2.5	NS
Suisun Bay off Chipps Island (508)	98	2.3	98	2.5	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.046	0.003	0.033	0.005	S*
High EC Control @ 19.88 mS/cm	0.023**	0.006	0.008**	0.001	S*
Rough and Ready DWR station, Stockton	0.095	0.005	0.105	0.006	NS
Montezuma Slough at Nurse Slough (609)	0.072	0.009	0.065	0.013	NS
Grizzly Bay at Dolphin (602)	0.044	0.007	0.049	0.003	NS
Napa River at Vallejo Seawall (340)[3]	0.052	0.004	0.032	0.007	S*
Carquinez Strait, West of Benicia army dock (405)	0.030*	0.007	0.044	0.008	NS
Suisun Bay off Chipps Island (508)	0.095	0.013	0.070	0.007	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard single-concentration statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

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

3. This high conductivity sample was compared to the High EC Control.

Table B 73-2. Summary of 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 4/14/09 - 4/15/09.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Rough and Ready DWR station, Stockton	914	16.9	7.72	8.0	10.2	0.15	0.002
Montezuma Slough at Nurse Slough (609)	3895	15.4	7.05	9.3	93.5	0.34	0.001
Grizzly Bay at Dolphin (602)	5620	15.6	7.54	10.0	234.0	0.32	0.002
Napa River at Vallejo Seawall (340)	19420	14.0	6.99	10.2	46.3	0.59	0.001
Carquinez Strait, West of Benicia army dock (405)	9520	14.3	7.32	10.1	146.0	0.62	0.002
Suisun Bay off Chipps Island (508)	597	14.9	7.14	10.2	26.8	0.31	0.001

Table B 73-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 4/16/09 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 4/14/09-4/15/09.

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 (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	331	22.0	23.4	7.6	8.4	7.73	8.02	100	56	-
High EC Control @ 19.88 mS/cm	19135	21.6	23.5	7.5	8.6	7.58	7.87	2680	82	-
Rough and Ready DWR station, Stockton	877	21.5	23.2	7.5	8.8	8.03	8.24	190	112	0.010
Montezuma Slough at Nurse Slough (609)	3577	20.2	23.4	7.4	8.4	7.82	8.01	452	88	0.013
Grizzly Bay at Dolphin (602)	6280	21.9	23.3	7.8	8.5	7.83	8.10	704	86	0.015
Napa River at Vallejo Seawall (340)	18650	22.6	23.2	6.4	8.1	7.56	7.91	2340	100	0.015
Carquinez Strait, West of Benicia army dock (405)	8875	22.7	23.1	7.4	8.5	7.80	8.02	1004	88	0.022
Suisun Bay off Chipps Island (508)	510	22.5	22.9	7.2	8.5	7.80	8.22	98	76	0.021
DIEPAMHR + 25 ppb PBO	338	22.5	22.5	7.7	8.6	7.83	8.12	-	-	-
High EC Control @ 19.88 mS/cm + 25 ppb PBO	19215	22.7	23.5	7.2	8.1	7.56	7.86	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	917	22.7	24.4	7.4	8.8	8.00	8.32	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	3754	22.6	24.4	7.3	8.7	7.83	8.03	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	7805	22.7	23.7	7.6	8.4	7.88	8.03	-	-	-
Napa River at Vallejo Seawall (340) + 25 ppb PBO	18885	22.6	24.8	7.1	8.5	7.77	7.95	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	8820	22.7	24.2	7.6	8.4	7.79	7.97	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	524	22.6	24.1	7.1	8.6	7.82	8.21	-	-	-

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 B 74-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 4/17/09 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 4/15/09 - 4/16/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	100	0.0	100	0.0	NS
Low EC control @147.3 uS/cm	100	0.0	94	3.2	NS
Sacramento R. Deep Water Channel, Light 55[3]	100	0.0	100	0.0	NS
Upper Cache Slough at mouth of Ulatis Creek	95	5.0	93	2.5	NS
Confluence of Linsey Sl. And Cache Sl.	98	2.5	95	2.9	NS
Suisun Slough at Rush Ranch	100	0.0	98	2.5	NS
Napa River at River Park Blvd.	100	0.0	95	2.9	NS
Sacramento River at tip of Grand Island (711)	95	2.8	93	4.8	NS
Sacramento River at Hood DWR Station[3]	97	2.8	95	2.9	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.065	0.006	0.056	0.001	NS
Low EC control @147.3 uS/cm	0.049*	0.005	0.033**	0.005	NS
Sacramento R. Deep Water Channel, Light 55[3]	0.092	0.010	0.092	0.015	NS
Upper Cache Slough at mouth of Ulatis Creek	0.101	0.011	0.096	0.009	NS
Confluence of Linsey Sl. And Cache Sl.	0.083	0.010	0.083	0.007	NS
Suisun Slough at Rush Ranch	0.050	0.014	0.090	0.005	S* (180%)
Napa River at River Park Blvd.	0.088	0.009	0.093	0.014	NS
Sacramento River at tip of Grand Island (711)	0.081	0.014	0.086	0.015	NS
Sacramento River at Hood DWR Station[3]	0.075	0.016	0.079	0.006	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

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

3. These low EC samples were compared to the Low EC Control.

Table B 74-2. Summary of 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 4/15/09 - 4/16/09.

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	409	15.5	7.23	9.5	18.0	0.14	0.001
Upper Cache Slough at mouth of Ulatis Creek	674	15.3	8.61	11.0	46.1	0.08	0.007
Confluence of Linsey Sl. And Cache Sl.	371	15.3	7.23	11.0	30.0	0.16	0.001
Suisun Slough at Rush Ranch	4816	16.9	7.53	8.8	395.3	0.46	0.004
Napa River at River Park Blvd.	3892	17.9	7.98	9.9	12.7	0.09	0.002
Sacramento River at tip of Grand Island (711)	148	15.6	7.03	10.0	23.9	0.42	0.001
Sacramento River at Hood DWR Station	148	14.4	6.85	10.0	7.8	0.52	0.001

Table B 74-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 4/17/09 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 4/15/09 - 4/16/09.

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 (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	326	22.7	23.4	7.7	8.2	7.81	8.01	104	60	-
Low EC control @147.3 uS/cm	155	22.6	23.2	7.6	8.9	7.55	7.85	44	26	-
Sacramento R. Deep Water Channel, Light 55	402	22.8	23.9	7.6	8.8	8.06	8.17	124	108	0.007
Upper Cache Slough at mouth of Ulatis Creek	675	22.7	23.5	7.5	8.6	8.35	8.47	226	204	0.010
Confluence of Linsey Sl. And Cache Sl.	366	22.7	24.3	7.3	8.5	8.00	8.20	114	118	0.009
Suisun Slough at Rush Ranch	4797	22.7	23.9	7.4	8.7	7.90	8.40	650	248	0.014
Napa River at River Park Blvd.	3703	22.8	23.9	7.6	8.5	7.97	8.16	480	120	0.003
Sacramento River at tip of Grand Island (711)	154	22.7	23.7	7.4	8.9	7.71	7.95	56	58	0.018
Sacramento River at Hood DWR Station	151	22.7	24.1	7.4	8.8	7.71	7.90	52	52	0.015
DIEPAMHR + 25 ppb PBO	346	22.7	23.6	7.6	8.5	7.80	8.01	-	-	-
Low EC control @147.3 uS/cm 25 ppb PBO	156	22.8	23.4	7.6	8.9	7.55	7.81	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	405	22.7	23.9	7.7	8.8	8.13	8.40	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	677	22.7	23.6	7.5	8.7	8.34	8.49	-	-	-
Confluence of Linsey Sl. And Cache Sl. + 25 ppb PBO	369	22.7	24.1	7.4	8.7	8.10	8.23	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	4806	22.7	23.7	7.4	8.7	7.78	8.39	-	-	-
Napa River at River Park Blvd. + 25 ppb PBO	3698	22.8	24.1	7.6	8.4	7.96	8.13	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	155	23.1	23.8	7.4	8.7	7.78	7.93	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	153	23.2	23.9	7.4	8.9	7.68	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 B 75-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 4/24/09 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 4/23/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	95	2.9	98	2.5	NS
Low EC Control @ 191.2 µS/cm	89	0.3	98	2.5	NS
Old River at mouth of Holland Cut (915)	90	10.0	100	0.0	NS
San Joaquin River at Potato Slough (815)	95	2.9	97	2.8	NS
Old River, western arm at railroad bridge (902)	100	0.0	95	2.9	NS
Field Dup: Old River, western arm at railroad bridge (902)	98	2.5	-	-	NA
Bottle Blank: DIEPAMHR	84	5.2	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.084	0.005	0.025	0.005	S*** (30%)
Low EC Control @ 191.2 µS/cm	0.061	0.004	0.031	0.002	S*** (51%)
Old River at mouth of Holland Cut (915)	0.087	0.007	0.058	0.007	S* (67%)
San Joaquin River at Potato Slough (815)	0.069	0.007	0.054	0.005	NS
Old River, western arm at railroad bridge (902)	0.068	0.004	0.045	0.003	S** (66%)
Field Dup: Old River, western arm at railroad bridge (902)	0.061	0.007	-	-	NA
Bottle Blank: DIEPAMHR	0.057**	0.003	-	-	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 (% non-PBO mean), NA: Not applicable.

*: $P < 0.05$

**: $P < 0.01$

***: $P < 0.001$

Table B 75-2. Summary of 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 4/23/09.

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 at Potato Slough (815)	181	18.2	7.16	9.9	7.0	0.10	0.000
Old River, western arm at railroad bridge (902)	252	20.5	7.24	9.4	6.8	0.06	0.000
Old River at mouth of Holland Cut (915)	292	20.5	7.19	9.0	6.4	0.03	0.000
Field Dup: Old River, western arm at railroad bridge (902)	252	20.5	7.24	9.4	7.4	0.06	0.000

Table B 75-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 4/24/09 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 4/23/09.

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 (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	339	22.7	24.4	7.4	8.5	7.80	8.11	-	-	-
Low EC Control @ 191.2 µS/cm	209	22.7	24.1	7.5	8.7	7.64	7.98	64	34	-
Old River at mouth of Holland Cut (915)	230	22.8	24.4	7.3	8.9	7.87	8.09	84	72	0.002
San Joaquin River at Potato Slough (815)	187	22.7	24.2	7.3	8.5	7.78	8.03	72	64	0.005
Old River, western arm at railroad bridge (902)	243	22.6	24.3	7.5	8.4	7.88	8.10	84	70	0.004
Field Dup: Old River, western arm at railroad bridge (902)	244	22.5	24.4	7.3	8.8	7.83	8.10	80	70	0.001
Bottle Blank: DIEPAMHR	334	22.6	24.2	7.5	8.6	7.76	8.08	64	58	0.003
DIEPAMHR + 25 ppb PBO	335	22.7	24.2	7.4	8.4	7.79	8.10	-	-	-
Low EC Control @ 191.2 µS/cm + 25 ppb PBO	209	22.7	24.2	7.5	8.9	7.69	7.96	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	283	22.8	24.1	7.3	8.9	7.73	8.06	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	184	22.7	24.1	7.2	8.9	7.74	7.96	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	245	22.6	24.0	7.4	8.6	7.83	8.13	-	-	-

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 B 76-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 4/30/09 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 4/28/09 - 4/29/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	98	2.5	94	3.2	NS
Low EC Control @ 129.1 µS/cm	98	2.5	98	2.5	NS
High EC Control @ 15.30 mS/cm	95	2.9	100	0.0	NS
High EC Control @ 25.00 mS/cm	79*	4.8	82	7.7	NS
Suisun Slough at Rush Ranch	100	0.0	100	0.0	NS
Napa River at River Park Blvd.	100	0.0	97	2.8	NS
Sacramento River at Hood DWR Station[3]	100	0.0	92	5.3	NS
Rough and Ready DWR station, Stockton	100	0.0	100	0.0	NS
Carquinez Strait, West of Benicia army dock (405)[4]	100	0.0	98	2.5	NS
Suisun Bay off Chipps Island (508)	98	2.3	100	0.0	NS
Montezuma Slough at Nurse Slough (609)	97	2.8	100	0.0	NS
Grizzly Bay at Dolphin (602)	98	2.5	95	2.8	NS
Napa River at Vallejo Seawall (340)[5]	90	5.5	95	3.1	NS
Trip Blank: DIEPAMHR	100	0.0	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.055	0.009	0.069	0.006	NS
Low EC Control @ 129.1 µS/cm	0.055	0.002	0.057	0.006	NS
High EC Control @ 15.30 mS/cm	0.036	0.006	0.045*	0.005	NS
High EC Control @ 25.00 mS/cm	0.020	0.005	0.034*	0.012	NS
Suisun Slough at Rush Ranch	0.090	0.006	0.119	0.006	S* (132%)
Napa River at River Park Blvd.	0.087	0.015	0.107	0.009	NS
Sacramento River at Hood DWR Station[3]	0.077	0.005	0.099	0.005	S* (129%)
Rough and Ready DWR station, Stockton	0.100	0.009	0.120	0.009	NS
Carquinez Strait, West of Benicia army dock (405)[4]	0.061	0.003	0.069	0.004	NS
Suisun Bay off Chipps Island (508)	0.098	0.002	0.106	0.011	NS
Montezuma Slough at Nurse Slough (609)	0.113	0.014	0.099	0.007	NS
Grizzly Bay at Dolphin (602)	0.054	0.007	0.081	0.004	S* (150%)
Napa River at Vallejo Seawall (340)[5]	0.048	0.007	0.070	0.005	S* (146%)
Trip Blank: DIEPAMHR	0.068	0.008	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard statistical protocols.
*: $P < 0.05$
2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.
3. This low conductivity sample was compared to the Low EC Control.
4. This high conductivity sample was compared to the High EC Control @ 15.30 mS/cm.
5. This high conductivity sample was compared to the High EC Control @ 25.00 mS/cm.

Table B 76-2. Summary of 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 4/28/09 - 4/29/09.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Suisun Slough at Rush Ranch	4772	14.7	7.01	8.9	71.4	0.14	0.000
Napa River at River Park Blvd.	9100	17.5	7.27	10.0	33.1	0.00	0.000
Sacramento River at Hood DWR Station	120	16.4	7.08	8.7	12.7	0.02	0.000
Rough and Ready DWR station, Stockton	690	18.8	7.74	7.5	12.6	0.13	0.002
Carquinez Strait, West of Benicia army dock (405)	15240	15.0	7.55	10.0	424.3	0.37	0.002
Suisun Bay off Chipps Island (508)	4810	15.4	7.51	10.0	37.0	0.13	0.001
Montezuma Slough at Nurse Slough (609)	4000	16.5	7.01	9.3	119.7	0.17	0.000
Grizzly Bay at Dolphin (602)	8380	15.3	7.48	10.0	379.0	0.33	0.002
Napa River at Vallejo Seawall (340)	24360	14.4	7.49	9.7	57.3	0.11	0.001
Trip Blank: DIEPAMHR	-	-	-	-	0.4	0.00	-

Table B 76-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 4/30/09 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 4/28/09 - 4/29/09.

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	332	22.8	24.1	7.3	8.2	7.70	8.26	124	58	-
Low EC Control @ 129.1 µS/cm	132	22.8	23.6	7.2	8.6	7.43	8.20	36	20	-
High EC Control @ 15.30 mS/cm	14790	22.9	23.9	7.2	8.3	7.68	7.99	1680	70	-
High EC Control @ 25.00 mS/cm	24215	22.8	24.0	7.1	7.8	7.72	7.93	3080	80	-
Suisun Slough at Rush Ranch	4586	22.8	24.0	7.5	8.4	8.11	8.15	640	158	0.007
Napa River at River Park Blvd.	4892	22.7	24.0	7.1	8.6	7.79	7.96	1200	128	0.000
Sacramento River at Hood DWR Station	235	22.7	24.1	7.0	8.7	7.62	8.11	48	51	0.001
Rough and Ready DWR station, Stockton	863	22.7	23.6	7.2	8.5	7.80	8.07	144	93	0.007
Carquinez Strait, West of Benicia army dock (405)	14030	22.7	22.8	7.2	8.2	7.68	7.88	1720	88	0.009
Suisun Bay off Chipps Island (508)	4674	22.7	23.9	7.4	8.7	7.71	8.01	520	74	0.005
Montezuma Slough at Nurse Slough (609)	3830	22.8	23.4	7.4	8.3	7.85	7.93	500	86	0.006
Grizzly Bay at Dolphin (602)	7910	22.9	23.8	7.4	8.2	7.75	8.07	1000	82	0.014
Napa River at Vallejo Seawall (340)	22870	22.8	23.8	6.9	7.6	7.64	7.87	2880	102	0.003
Trip Blank: DIEPAMHR	446	22.9	23.9	7.3	8.6	7.73	8.16	100	57	0.000
DIEPAMHR + 25 ppb PBO	339	22.5	22.8	7.3	8.4	7.70	8.03	-	-	-
Low EC Control @ 129.1 µS/cm + 25 ppb PBO	161	22.5	22.8	7.2	8.5	7.44	8.12	-	-	-
High EC Control @ 15.30 mS/cm + 25 ppb PBO	14580	22.4	22.9	7.3	8.2	7.67	7.91	-	-	-
High EC Control @ 25.00 mS/cm + 25 ppb PBO	23910	22.5	22.9	7.0	8.0	7.75	7.96	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	4550	22.4	22.9	7.4	8.6	7.92	8.21	-	-	-
Napa River at River Park Blvd. + 25 ppb PBO	4820	22.3	22.9	7.3	8.8	7.86	7.97	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	166	22.2	22.9	7.1	8.6	7.52	8.16	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	699	22.1	22.8	7.3	8.6	7.82	8.01	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	14075	22.0	22.8	7.1	8.1	7.72	7.91	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	4428	21.9	22.8	7.4	8.9	7.72	7.98	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	3710.5	21.8	22.8	7.2	8.3	7.81	7.93	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	7890	21.9	22.9	7.5	8.3	7.74	7.94	-	-	-
Napa River at Vallejo Seawall (340) + 25 ppb PBO	22215	21.4	22.8	6.7	8.4	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 B 77-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 5/01/09 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 4/30/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	98	2.5	98	2.5	NS
Low EC Control @ 120.5 uS/cm	97	2.8	100	0.0	NS
Sacramento R. Deep Water Channel, Light 55	100	0.0	98	2.5	NS
Upper Cache Slough at mouth of Ulatis Creek	100	0.0	100	0.0	NS
Confluence of Lindsey Sl. and Cache Sl.	100	0.0	98	2.5	NS
Sacramento River at tip of Grand Island (711)	97	2.8	98	2.5	NS
San Joaquin River at Potato Slough	98	2.5	100	0.0	NS
Old River, western arm at railroad bridge (902)	100	0.0	100	0.0	NS
Old River at mouth of Holland Cut (915)	100	0.0	98	2.5	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.057	0.004	0.048	0.001	S*
Low EC Control @ 120.5 uS/cm	0.051	0.006	0.025**	0.004	S**
Sacramento R. Deep Water Channel, Light 55	0.095	0.005	0.090	0.004	NS
Upper Cache Slough at mouth of Ulatis Creek	0.083	0.009	0.087	0.005	NS
Confluence of Lindsey Sl. and Cache Sl.	0.106	0.006	0.085	0.009	S*
Sacramento River at tip of Grand Island (711)	0.092	0.008	0.093	0.005	NS
San Joaquin River at Potato Slough	0.105	0.006	0.114	0.010	NS
Old River, western arm at railroad bridge (902)	0.109	0.006	0.112	0.009	NS
Old River at mouth of Holland Cut (915)	0.118	0.010	0.116	0.003	NS

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

*: $P < 0.05$

**: $P < 0.01$

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

Table B 77-2. Summary of 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 4/30/09.

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	236	16.6	7.25	9.4	31.2	0.16	0.001
Upper Cache Slough at mouth of Ulatis Creek	329	16.8	6.88	9.9	45.9	0.03	0.000
Confluence of Lindsey Sl. and Cache Sl.	246	16.8	6.80	9.5	27.5	0.20	0.000
Sacramento River at tip of Grand Island (711)	120	17.1	6.88	9.8	10.1	0.04	0.000
San Joaquin River at Potato Slough (815)	196	19.0	6.82	10.0	4.4	0.07	0.000
Old River, western arm at railroad bridge (902)	243	18.3	6.81	9.3	6.1	0.04	0.000
Old River at mouth of Holland Cut (915)	294	19.2	6.86	9.3	5.9	0.02	0.000

Table B 77-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 5/1/09 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 4/30/09.

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 (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	337	23.3	23.8	7.4	8.3	7.66	8.10	124	58	-
Low EC Control @ 120.5 uS/cm	125	23.2	23.8	7.2	8.6	7.29	8.05	44	20	-
Sacramento R. Deep Water Channel, Light 55	261	23.3	23.7	7.1	8.7	7.75	8.13	84	78	0.010
Upper Cache Slough at mouth of Ulatis Creek	320	23.4	23.9	7.3	8.3	7.95	8.24	100	102	0.002
Confluence of Lindsey Sl. and Cache Sl.	203	23.3	23.7	7.2	8.9	7.74	8.19	64	74	0.014
Sacramento River at tip of Grand Island (711)	120	23.3	23.8	6.9	8.4	7.52	8.11	48	46	0.002
San Joaquin River at Potato Slough (815)	195	23.1	23.7	7.2	8.8	7.73	8.14	64	66	0.004
Old River, western arm at railroad bridge (902)	240	23.3	23.9	7.3	8.5	7.75	8.20	56	74	0.003
Old River at mouth of Holland Cut (915)	289	23.3	23.7	7.2	8.4	7.79	8.17	88	78	0.001
DIEPAMHR + 25 ppb PBO	335	22.0	23.9	7.4	8.5	7.70	8.06	-	-	-
Low EC Control @ 120.5 uS/cm + 25 ppb PBO	117	22.0	23.6	7.2	8.5	7.29	7.95	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	245	22.2	23.8	7.1	8.4	7.72	8.13	-	-	-
Upper Cache Slough at mouth of Ulatis Creek (815) + 25 ppb PBO	303	21.8	23.7	7.2	8.6	7.97	8.29	-	-	-
Confluence of Lindsey Sl. and Cache Sl. + 25 ppb PBO	189	21.7	23.9	7.3	8.6	7.75	8.14	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	112	21.8	23.7	7.0	8.6	7.57	8.10	-	-	-
San Joaquin River at Potato Slough + 25 ppb PBO	184	21.8	23.9	7.3	8.9	7.74	7.99	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	231	21.8	23.9	7.3	8.6	7.78	8.09	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	275	22.0	24.0	7.3	8.7	7.77	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 B 78-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 5/14/09 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 5/12/09-5/13/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	95	5.0	-	-	-
Low EC Control @ 119.2 µS	90	5.8	-	-	-
High EC Control @ 17.30 mS	78	11.9	-	-	-
Napa River, near River Park Blvd.	98	2.5	-	-	-
Suisun Slough at Rush Ranch	95	2.8	-	-	-
Sacramento River at Hood DWR Station	72	8.7	-	-	-
Rough and Ready DWR station, Stockton	98	2.5	-	-	-
Napa River at Vallejo Seawall (340)	57	4.1	-	-	-

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.081	0.008	-	-	-
Low EC Control @ 119.2 µS	0.053	0.009	-	-	-
High EC Control @ 17.30 mS	0.051	0.004	-	-	-
Napa River, near River Park Blvd.	0.067	0.011	-	-	-
Suisun Slough at Rush Ranch	0.072	0.008	-	-	-
Sacramento River at Hood DWR Station	0.054	0.008	-	-	-
Rough and Ready DWR station, Stockton	0.095	0.003	-	-	-
Napa River at Vallejo Seawall (340)	0.099	0.021	-	-	-

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.

* This test exhibited contamination in the PBO-manipulated samples, due to contamination of the PBO stock solution. This test was reset up on 5/16/09.

Table B 78-2. Summary of 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 5/12/09-5/13/09.

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, near River Park Blvd.	5780	20.8	6.51	10.6	47.8	0.14	0.005
Suisun Slough at Rush Ranch	4863	19.0	6.51	9.8	62.3	0.32	0.017
Sacramento River at Hood DWR Station	116	19.3	6.89	11.4	21.5	0.21	0.010
Rough and Ready DWR station, Stockton	491	21.3	7.43	6.9	9.6	0.09	0.003
Napa River at Vallejo Seawall (340)	16330	17.5	6.91	9.2	77.5	0.13	0.003

Table B 78-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 5/14/09 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 5/12/09-5/13/09.

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	22.9	23.4	7.6	8.5	7.71	8.07	108	60	-
Low EC Control @ 119.2 µS	122	23.0	23.3	7.5	8.6	7.27	7.91	32	22	-
High EC Control @ 17.30 mS	16615	23.1	23.9	7.2	8.7	7.66	7.85	2040	82	-
Napa River, near River Park Blvd.	5530	23.0	23.6	6.9	8.4	7.86	8.01	652	116	0.004
Suisun Slough at Rush Ranch	4675	23.1	23.8	6.9	8.3	7.91	8.18	584	198	0.010
Sacramento River at Hood DWR Station	131	23.4	23.6	6.9	8.5	7.48	7.82	44	50	0.007
Rough and Ready DWR station, Stockton	477	23.2	23.8	7.0	8.5	7.67	8.04	112	70	0.004
Napa River at Vallejo Seawall (340)	15380	23.1	23.6	6.9	8.2	7.67	7.89	1920	94	0.008

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 B 79-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 5/15/09 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 5/14/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	91	5.9	58	25.0	NS
Confluence of Lindsey Sl. and Cache Sl.	95	2.9	87	9.4	NS
Sacramento R. Deep Water Channel, Light 55	92	5.3	100	0.0	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.073	0.005	0.076	0.008	NS
Confluence of Lindsey Sl. and Cache Sl.	0.090	0.007	0.089	0.010	NS
Sacramento R. Deep Water Channel, Light 55	0.098	0.005	0.109	0.008	NS

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

Table B 79-2. Summary of 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 5/14/09.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (^oC)	pH	DO (mg/L)			
Confluence of Lindsey Sl. and Cache Sl.	207	20.7	7.29	11.0	132.7	0.16	0.001
Sacramento R. Deep Water Channel, Light 55	261	21.8	7.82	8.8	96.9	0.17	0.005

Table B 79-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 5/15/09 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 5/14/09.

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 (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	359	23.4	24.3	7.4	8.6	7.70	8.00	80	58	-
Confluence of Lindsey Sl. and Cache Sl.	254	23.5	24.2	7.3	8.6	7.70	8.17	76	74	0.011
Sacramento R. Deep Water Channel, Light 55	291	23.6	24.2	7.4	8.5	7.61	8.03	76	72	0.009
DIEPAMHR + 25 ppb PBO	362	23.4	24.0	7.3	8.5	7.72	8.02	-	-	-
Confluence of Lindsey Sl. and Cache Sl. + 25 ppb PBO	245	23.9	24.0	7.2	8.7	7.72	8.18	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	290	24.0	24.3	7.1	8.7	7.69	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 B 80-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 5/16/09 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 5/12/09 - 5/13/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	100	0.0	87	9.4	NS
Low EC Control @ 119.2 µS/cm	100	0.0	87	6.3	NS
High EC Control @ 17.30 mS/cm	73*	6.0	73	11.1	NS
Napa River at River Park Blvd.	100	0.0	98	2.5	NS
Suisun Slough at Rush Ranch	100	0.0	97	2.8	NS
Sacramento River at Hood DWR Station[3]	90	4.1	81	3.3	NS
Rough and Ready DWR station, Stockton	100	0.0	100	0.0	NS
Napa River at Vallejo Seawall (340)[4]	61	10.1	14**	9.0	S*

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.062	0.006	0.047	0.003	S*
Low EC Control @ 119.2 µS/cm	0.049	0.006	0.045	0.006	NS
High EC Control @ 17.30 mS/cm	0.039*	0.005	0.027**	0.004	NS
Napa River at River Park Blvd.	0.099	0.007	0.069	0.002	S**
Suisun Slough at Rush Ranch	0.101	0.006	0.089	0.012	NS
Sacramento River at Hood DWR Station[3]	0.070	0.008	0.043	0.007	S*
Rough and Ready DWR station, Stockton	0.085	0.004	0.084	0.007	NS
Napa River at Vallejo Seawall (340)[4]	0.063	0.006	0.097	0.013	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

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

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

4. This high conductivity sample was compared to the High EC Control.

Table B 80-2. Summary of 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 5/12/09 - 5/13/09.

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 River Park Blvd.	5780	20.8	6.51	10.6	47.8	0.14	0.000
Suisun Slough at Rush Ranch	4863	19.0	6.51	9.8	62.3	0.32	0.000
Sacramento River at Hood DWR Station	116	19.3	6.89	11.4	21.5	0.21	0.001
Rough and Ready DWR station, Stockton	491	21.3	7.43	6.9	9.6	0.09	0.001
Napa River at Vallejo Seawall (340)	16330	17.5	6.91	9.2	77.5	0.13	0.000

Table B 80-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 5/16/09 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 5/12/09 - 5/13/09.

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	23.9	24.0	7.5	8.5	7.72	8.06	108	60	-
Low EC Control @ 119.2 µS/cm	127	23.7	24.1	7.4	8.5	7.30	7.82	32	22	-
High EC Control @ 17.30 mS/cm	16505	23.9	24.1	6.9	8.1	7.56	7.82	2040	82	-
Napa River at River Park Blvd.	5410	23.8	24.0	6.9	8.2	7.78	7.98	652	116	0.005
Suisun Slough at Rush Ranch	4550	23.9	24.0	6.9	8.5	8.01	8.19	384	198	0.017
Sacramento River at Hood DWR Station	124	23.7	24.0	7.0	8.4	7.34	7.99	44	50	0.010
Rough and Ready DWR station, Stockton	483	23.9	24.1	7.2	8.4	7.66	7.91	112	70	0.003
Napa River at Vallejo Seawall (340)	15320	23.7	24.0	6.6	8.1	7.58	7.81	1920	94	0.003
DIEPAMHR + 25 ppb PBO	239	23.7	23.7	7.2	8.5	7.67	7.84	-	-	-
Low EC Control @ 119.2 µS/cm + 25 ppb PBO	240	23.6	24.0	7.4	8.5	7.37	8.08	-	-	-
High EC Control @ 17.30 mS/cm + 25 ppb PBO	16445	23.9	24.0	6.7	8.0	7.60	7.84	-	-	-
Napa River at River Park Blvd. + 25 ppb PBO	5330	23.8	24.0	7.2	8.4	7.78	7.98	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	4552	23.8	24.3	6.9	8.3	7.99	8.20	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	149	23.8	24.2	6.9	8.7	7.44	7.83	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	485	23.9	24.2	7.0	8.5	7.65	7.94	-	-	-
Napa River at Vallejo Seawall	15325	23.6	24.1	6.8	8.2	7.68	7.78	-	-	-

(340) + 25 ppb PBO

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 B 81-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 5/20/09 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 5/18/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	98	2.5	100	0.0	NS
High EC Control @ 20360 uS/cm	100	0.0	93	4.8	NS
Grizzly Bay at Dolphin (602)	100	0.0	100	0.0	NS
Suisun Bay off Chipps Island (508)	100	0.0	100	0.0	NS
Carquinez Strait, West of Benicia army dock (405)[3]	88	7.5	93	4.8	NS
Montezuma Slough at Nurse Slough (609)	100	0.0	100	0.0	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.071	0.002	0.081	0.005	NS
High EC Control @ 20360 uS/cm	0.035*	0.012	0.051**	0.005	NS
Grizzly Bay at Dolphin (602)	0.096	0.002	0.103	0.007	NS
Suisun Bay off Chipps Island (508)	0.087	0.008	0.111	0.010	NS
Carquinez Strait, West of Benicia army dock (405)[3]	0.053	0.007	0.058	0.002	NS
Montezuma Slough at Nurse Slough (609)	0.106	0.009	0.102	0.004	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

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

3. This high conductivity sample was compared to the High EC Control.

Table B 81-2. Summary of 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 5/18/09.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (^{o}C)	pH	DO (mg/L)			
Grizzly Bay at Dolphin (602)	6250	19.7	7.95	9.0	54.5	0.09	0.003
Suisun Bay off Chipps Island (508)	2366	19.5	6.98	9.0	28.3	0.00	0.000
Carquinez Strait, West of Benicia army dock (405)	19550	18.3	7.38	9.3	10.5	0.00	0.000
Montezuma Slough at Nurse Slough (609)	3368	21.4	7.66	8.2	47.0	0.00	0.000

Table B 81-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 5/20/09 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 5/18/09.

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 (^{o}C)	Max Temp (^{o}C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	340	22.9	23.5	7.4	8.4	7.74	8.20	104	60	-
High EC Control @ 20360 uS/cm	19550	23.6	23.6	7.0	8.4	7.63	7.85	2400	90	-
Grizzly Bay at Dolphin (602)	6135	23.0	23.6	7.4	8.4	7.63	7.97	760	76	0.003
Suisun Bay off Chipps Island (508)	2237	23.4	23.6	7.5	8.8	7.65	8.06	256	64	0.000
Carquinez Strait, West of Benicia army dock (405)	18545	23.4	23.6	7.0	8.4	7.66	7.84	2200	94	0.000
Montezuma Slough at Nurse Slough (609)	3159	23.5	23.7	6.8	8.5	7.64	8.07	360	84	0.000
DIEPAMHR + 25 ppb PBO	331	23.1	23.4	7.3	8.4	7.72	8.05	-	-	-

High EC Control @ 20360 uS/cm + 25 ppb PBO	19385	23.2	23.5	6.9	8.0	7.58	7.78	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	5685	23.0	23.9	7.3	8.3	7.66	7.90	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	2217	23.0	24.0	7.1	8.9	7.68	7.97	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	18285	23.0	24.4	7.0	8.9	7.65	7.82	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	3173	23.2	23.6	7.4	8.5	7.69	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 B 82-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 5/21/09 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 5/20/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	97	2.8	97	2.8	NS
Low EC Control @ 149.4 µS/cm	98	2.5	95	2.9	NS
Upper Cache Slough at mouth of Ulatis Creek	100	0.0	100	0.0	NS
Sacramento River at tip of Grand Island (711)	100	0.0	93	4.8	NS
San Joaquin River at Potato Slough (815)	100	0.0	100	0.0	NS
Old River, western arm at railroad bridge (902)	100	0.0	100	0.0	NS
Old River at mouth of Holland Cut (915)	100	0.0	100	0.0	NS
Bottle Blank (amber cubitainer)	100	0.0	-	-	NA
Bottle Blank (clear cubitainer)	100	0.0	-	-	NA

Treatment	Weight (mg/surviving individual)[1]

	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.061	0.008	0.077	0.010	NS
Low EC Control @ 149.4 µS/cm	0.073	0.006	0.065	0.006	NS
Upper Cache Slough at mouth of Ulatis Creek	0.102	0.007	0.097	0.004	NS
Sacramento River at tip of Grand Island (711)	0.091	0.008	0.089	0.003	NS
San Joaquin River at Potato Slough (815)	0.082	0.009	0.095	0.010	NS
Old River, western arm at railroad bridge (902)	0.078	0.005	0.097	0.005	NS
Old River at mouth of Holland Cut (915)	0.087	0.011	0.104	0.009	NS
Bottle Blank (amber cubitainer)	0.067	0.003	-	-	NA
Bottle Blank (clear cubitainer)	0.065	0.005	-	-	NA

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

Table B 82-2. Summary of 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 5/20/09.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Upper Cache Slough at mouth of Ulatis Creek	282	19.9	7.24	8.3	56.6	0.09	0.001
Sacramento River at tip of Grand Island (711)	144	20.7	6.61	8.4	8.0	0.38	0.001
San Joaquin River at Potato Slough (815)	205	21.8	6.58	8.4	7.5	0.10	0.000
Old River, western arm at railroad bridge (902)	229	21.8	6.58	8.4	6.6	0.08	0.000
Old River at mouth of Holland Cut (915)	320	22.7	6.43	8.3	4.7	0.03	0.000
Bottle Blank (amber cubitainer)	-	-	-	-	-	0.05	-
Bottle Blank (clear cubitainer)	-	-	-	-	-	0.03	-

Table B 82-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 5/21/09 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 5/20/09.

Treatment	Laboratory Chemistry	Hardness	Alkalinity	Unionized

	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH	(mg/L as $CaCO_3$)	(mg/L as $CaCO_3$)	Ammonia (mg/L)[1]
DIEPAMHR	334	23.0	24.5	7.4	8.6	7.68	8.03	104	60	-
Low EC Control @ 149.4 µS/cm	145	23.2	24.0	7.2	8.6	7.33	7.69	44	28	-
Upper Cache Slough at mouth of Ulatis Creek	236	23.0	24.5	7.1	8.8	7.83	8.19	108	92	0.006
Sacramento River at tip of Grand Island (711)	103	22.4	24.4	6.9	8.7	7.46	7.91	48	56	0.014
San Joaquin River at Potato Slough (815)	183	22.8	24.1	7.0	8.8	7.58	7.99	60	58	0.004
Old River, western arm at railroad bridge (902)	167	23.7	24.1	7.0	8.7	7.64	8.03	72	64	0.004
Old River at mouth of Holland Cut (915)	242	23.0	24.2	6.9	8.7	7.65	8.11	84	76	0.002
Bottle Blank (amber cubitainer)	261	22.9	24.8	7.3	8.7	7.75	8.05	104	60	0.002
Bottle Blank (clear cubitainer)	313	22.6	24.6	7.4	8.7	7.68	8.03	104	60	0.001
DIEPAMHR + 25 ppb PBO	329	22.5	24.4	7.4	8.5	7.71	8.06	-	-	-
Low EC Control @ 149.4 µS/cm + 25 ppb PBO	144	22.3	23.8	7.3	8.6	7.32	7.76	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	235	22.1	24.5	7.1	8.6	7.81	8.18	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	134	22.1	24.4	6.8	8.6	7.46	7.93	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	221	22.2	24.4	7.1	8.6	7.60	7.96	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	205	22.6	25.1	7.0	8.8	7.62	8.04	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	280	22.1	25.5	7.0	8.8	7.74	8.07	-	-	-

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 B 83-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 5/28/09 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 5/26/09 - 5/27/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	95	3.1	100	0.0	NS
Low EC Control @ 157.5 µS/cm	100	0.0	100	0.0	NS
High EC Control @ 14.50 mS/cm	100	0.0	98	2.5	NS
High EC Control @ 23.36 mS/cm	94	3.3	94	6.3	NS
Suisun Slough at Rush Ranch	100	0.0	100	0.0	NS
Napa River at River Park Blvd.	100	0.0	98	2.5	NS
Sacramento River at Hood DWR Station[3]	94	3.3	98	2.5	NS
Grizzly Bay at Dolphin (602)	97	2.8	98	2.5	NS
Suisun Bay off Chipps Island (508)	100	0.0	98	2.5	NS
Napa River at Vallejo Seawall (340)[5]	90	5.8	94	3.2	NS
Montezuma Slough at Nurse Slough (609)	98	2.5	98	2.5	NS
Carquinez Strait, West of Benicia army dock (405)[4]	98	2.5	100	0.0	NS
Bottle Blank: DIEPAMHR	100	0.0	-	-	NA
Field Dup.: Suisun Slough at Rush Ranch	98	2.5	-	-	NA

Field Dup.: Montezuma Slough at Nurse Slough (609)	100	0.0	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.035	0.006	0.078	0.006	S** (223%)
Low EC Control @ 157.5 µS/cm	0.036	0.005	0.045	0.008	NS
High EC Control @ 14.50 mS/cm	0.037	0.003	0.055	0.004	S** (149%)
High EC Control @ 23.36 mS/cm	0.037	0.006	0.036	0.007	NS
Suisun Slough at Rush Ranch	0.097	0.010	0.123	0.008	S* (127%)
Napa River at River Park Blvd.	0.093	0.011	0.084	0.006	NS
Sacramento River at Hood DWR Station[3]	0.087	0.006	0.090	0.008	NS
Grizzly Bay at Dolphin (602)	0.087	0.010	0.097	0.005	NS
Suisun Bay off Chipps Island (508)	0.080	0.003	0.098	0.005	S* (123%)
Napa River at Vallejo Seawall (340)[5]	0.048	0.004	0.055	0.005	NS
Montezuma Slough at Nurse Slough (609)	0.090	0.001	0.075	0.005	S* (83%)
Carquinez Strait, West of Benicia army dock (405)[4]	0.073	0.000	0.117	0.041	NS
Bottle Blank: DIEPAMHR	0.070	0.008	-	-	NA
Field Dup.: Suisun Slough at Rush Ranch	0.112	0.009	-	-	NA
Field Dup.: Montezuma Slough at Nurse Slough (609)	0.103	0.008	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard statistical protocols.
*: $P < 0.05$
**: $P < 0.01$
2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.
3. This low conductivity sample was compared to the Low EC Control.
4. This high conductivity sample was compared to the High EC Control @ 14.50 mS/cm.
5. This high conductivity sample was compared to the High EC Control @ 23.36 mS/cm.

Table B 83-2. Summary of 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 5/26/09 - 5/27/09.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Suisun Slough at Rush Ranch	4755	18.5	6.82	7.1	54.7	0.12	0.000
Napa River at River Park Blvd.	10530	20.1	6.86	8.5	13.0	0.11	0.000
Sacramento River at Hood DWR Station	152	20.7	6.55	8.3	15.3	0.33	0.000
Grizzly Bay at Dolphin (602)	8100	22.1	6.80	9.1	28.4	0.06	0.000
Suisun Bay off Chipps Island (508)	3924	19.5	6.54	9.1	36.9	0.09	0.000
Napa River at Vallejo Seawall (340)	22870	18.6	6.58	8.7	32.8	0.09	0.000
Montezuma Slough at Nurse Slough (609)	3446	21.1	6.66	8.1	137.7	0.16	0.000
Carquinez Strait, West of Benicia army dock (405)	14080	18.5	7.45	9.2	288.7	0.21	0.001
Field Dup.: Suisun Slough at Rush Ranch	4755	18.5	6.82	7.1	47.7	0.07	0.000
Field Dup.: Montezuma Slough at Nurse Slough (609)	3446	21.1	6.66	8.1	138.0	0.15	0.000

Table B 83-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 5/28/09 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 5/26/09 - 5/27/09.

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 (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	332	23.5	23.7	7.5	8.6	7.59	8.18	104	60	-
Low EC Control @ 157.5 µS/cm	157	23.6	23.6	7.6	8.8	7.42	7.97	52	26	-
High EC Control @ 14.50 mS/cm	14125	23.6	23.6	7.1	8.1	7.65	7.89	1720	74	-
High EC Control @ 23.36 mS/cm	22435	23.4	23.7	7.1	8.1	7.72	7.87	2760	90	-
Suisun Slough at Rush Ranch	4504	23.5	23.9	7.3	8.5	7.81	8.13	520	142	0.003
Napa River at River Park Blvd.	10125	23.4	23.7	7.0	8.6	7.84	7.98	1240	128	0.003
Sacramento River at Hood DWR Station	146	23.5	23.7	7.1	8.4	7.62	7.82	52	54	0.010
Grizzly Bay at Dolphin (602)	7690	23.4	23.7	7.3	8.4	7.67	7.92	960	68	0.002
Suisun Bay off Chipps Island (508)	4089	23.3	23.7	7.4	8.6	7.65	7.89	500	64	0.003
Napa River at Vallejo Seawall (340)	21675	23.2	23.8	6.8	8.3	7.67	7.81	2640	100	0.001
Montezuma Slough at Nurse Slough (609)	3253	23.2	23.8	7.4	8.4	7.80	7.94	420	92	0.005
Carquinez Strait, West of Benicia army dock (405)	13450	23.2	23.7	7.2	8.1	7.61	7.81	1600	94	0.005
Bottle Blank 052609	343	23.2	23.8	7.5	8.6	7.77	8.18	104	60	-
Field Dup.: Suisun Slough at Rush Ranch	4542	23.2	23.8	7.3	8.7	7.90	8.13	52	154	0.002
Field Dup.: Montezuma Slough at Nurse Slough (609)	3292	23.2	23.8	7.5	8.9	7.78	7.96	420	84	0.006
DIEPAMHR + 25 ppb PBO	339	23.1	23.5	7.5	8.7	7.73	8.24	-	-	-
Low EC Control @ 157.5 µS/cm + 25 ppb PBO	160	23.1	23.5	7.7	8.9	7.46	8.01	-	-	-
High EC Control @ 14.50 mS/cm + 25 ppb PBO	13795	23.0	23.6	7.0	8.4	7.59	7.90	-	-	-
High EC Control @ 23.36 mS/cm + 25 ppb PBO	22405	23.1	23.7	6.9	8.2	7.65	7.88	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	4537.5	23.0	23.8	7.5	8.8	7.81	8.14	-	-	-
Napa River at River Park Blvd. + 25 ppb PBO	10045	23.0	23.9	7.4	8.7	7.90	8.00	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	159.2	23.0	23.8	7.3	8.8	7.63	8.00	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	7665	23.1	24.0	7.4	8.9	7.65	7.80	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	4036	23.0	24.1	7.4	8.8	7.60	7.94	-	-	-
Napa River at Vallejo Seawall (340) + 25 ppb PBO	21460	23.1	24.0	6.8	8.4	7.68	7.79	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	3294.5	23.0	23.8	7.6	8.8	7.81	8.07	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	13630	23.0	23.9	7.12	8.6	7.61	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 B 84-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 5/29/09 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 5/27/09 and 5/28/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	98	2.5	100	0.0	NS
Low EC Control @ 139.6 µS/cm	95	3.1	98	2.5	NS
Rough and Ready DWR station, Stockton	100	0.0	98	2.5	NS
Sacramento R. Deep Water Channel, Light 55[3]	98	2.5	100	0.0	NS
Confluence of Lindsey Sl. and Cache Sl.[3]	98	2.5	100	0.0	NS
Upper Cache Slough at mouth of Ulatis Creek[3]	100	0.0	98	2.5	NS
Sacramento River at tip of Grand Island (711)[3]	93	2.5	98	2.5	NS
Old River at mouth of Holland Cut (915)	95	2.9	100	0.0	NS
San Joaquin River at Potato Slough (815)[3]	98	2.5	100	0.0	NS
Old River, western arm at railroad bridge (902)[3]	100	0.0	98	2.5	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.030	0.005	0.034	0.004	NS
Low EC Control @ 139.6 µS/cm	0.036	0.002	0.036	0.006	NS
Rough and Ready DWR station, Stockton	0.093	0.009	0.087	0.012	NS
Sacramento R. Deep Water Channel, Light 55[3]	0.082	0.008	0.075	0.011	NS
Confluence of Lindsey Sl. and Cache Sl.[3]	0.068	0.010	0.081	0.008	NS
Upper Cache Slough at mouth of Ulatis Creek[3]	0.082	0.003	0.083	0.006	NS
Sacramento River at tip of Grand Island (711)[3]	0.045	0.009	0.074	0.014	NS
Old River at mouth of Holland Cut (915)	0.094	0.009	0.094	0.006	NS
San Joaquin River at Potato Slough (815)[3]	0.079	0.030	0.085	0.009	NS
Old River, western arm at railroad bridge (902)[3]	0.091	0.011	0.100	0.013	NS

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

3. These low conductivity samples were compared to the Low EC Control.

Table B 84-2. Summary of 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 5/27/09 - 5/28/09.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (^{o}C)	pH	DO (mg/L)			
Rough and Ready DWR station, Stockton	435	25.8	7.08	6.0	13.3	0.09	0.001
Sacramento R. Deep Water Channel, Light 55	215	20.9	7.39	8.4	32.2	0.14	0.001
Confluence of Lindsey Sl. and Cache Sl.	188	21.1	7.52	8.4	37.8	0.11	0.001
Upper Cache Slough at mouth of Ulatis Creek	243	21.2	7.50	8.4	63.5	0.04	0.001
Sacramento River at tip of Grand Island (711)	151	21.8	7.54	8.4	11.6	0.22	0.003
Old River at mouth of Holland Cut (915)	286	24.1	7.18	7.7	6.4	0.00	0.000
San Joaquin River at Potato Slough (815)	176	22.7	7.27	8.2	6.3	0.00	0.000
Old River, western arm at railroad bridge (902)	231	23.6	7.29	8.0	7.2	0.00	0.000

Table B 84-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 5/29/09 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 5/27/09 - 5/28/09.

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 (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	339	23.1	23.5	7.3	8.4	7.66	8.08	108	62	-
Low EC Control @ 139.6 µS/cm	145	23.3	23.6	7.2	8.8	7.37	7.83	40	25	-
Rough and Ready DWR station, Stockton	422	23.3	23.5	7.0	8.7	7.70	7.82	96	68	0.002
Sacramento R. Deep Water Channel, Light 55	194	23.3	23.6	6.9	8.7	7.70	7.94	64	66	0.006
Confluence of Lindsey Sl. and Cache Sl.	177	23.3	23.5	6.6	8.7	7.70	7.96	64	62	0.005
Upper Cache Slough at mouth of Ulatis Creek	229	23.3	23.5	6.4	8.9	7.75	8.00	76	80	0.002
Sacramento River at tip of Grand Island (711)	145	23.3	23.5	6.9	8.5	7.62	7.75	52	54	0.006
Old River at mouth of Holland Cut (915)	271	23.3	23.5	6.3	8.6	7.80	7.87	84	70	0.000
San Joaquin River at Potato Slough (815)	172	23.3	23.5	6.4	8.8	7.62	7.92	56	52	0.000
Old River, western arm at railroad bridge (902)	214	23.3	23.5	6.3	8.8	7.69	7.94	68	60	0.000
DIEPAMHR + 25 ppb PBO	338	23.4	23.5	7.0	8.8	7.65	8.09	-	-	-
Low EC Control @ 139.6 µS/cm + 25 ppb PBO	146	23.4	23.5	6.7	8.6	7.31	7.84	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	423	23.3	23.5	6.7	8.4	7.71	7.80	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	197	23.5	23.5	6.4	8.8	7.69	7.93	-	-	-
Confluence of Lindsey Sl. and Cache Sl. + 25 ppb PBO	181	23.4	23.5	6.6	8.6	7.70	7.96	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	232	23.4	23.5	6.4	8.8	7.77	8.10	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	144	23.6	23.9	6.3	8.4	7.61	8.00	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	266	23.6	23.7	6.2	8.9	7.77	8.40	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	164	23.7	23.7	6.2	8.5	7.61	7.90	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	209	23.6	23.8	6.1	8.7	7.74	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 B 85-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 6/11/09 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 6/09/09 - 6/10/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	95	2.9	98	2.5	NS
Low EC Control @ 161.5 µS/cm	90	7.1	95	3.1	NS
High EC Control @ 14.06 mS/cm	95	3.1	95	2.9	NS
High EC Control @ 23.81 mS/cm	73	4.8	78	3.9	NS
Suisun Slough at Rush Ranch	100	0.0	100	0.0	NS
Sacramento River at Hood DWR Station[3]	87	4.7	74	6.6	NS
Napa River at River Park Blvd.[4]	98	2.5	95	2.8	NS
Rough and Ready DWR station, Stockton	100	0.0	95	5.0	NS
Napa River at Vallejo Seawall (340)[5]	74	1.6	91	5.4	NS
Montezuma Slough at Nurse Slough (609)	100	0.0	98	2.5	NS
Suisun Bay off Chipps Island (508)	100	0.0	100	0.0	NS
Grizzly Bay at Dolphin (602)	97	2.8	100	0.0	NS
Carquinez Strait, West of Benicia army dock (405)[4]	100	0.0	98	2.5	NS
Trip Blank: DIEPAMHR	97	3.1	-	-	NA
Bottle Blank: DIEPAMHR	92	2.6	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.053	0.009	0.042	0.004	NS
Low EC Control @ 161.5 µS/cm	0.044	0.003	0.036	0.007	NS
High EC Control @ 14.06 mS/cm	0.048	0.006	0.045	0.008	NS
High EC Control @ 23.81 mS/cm	0.033	0.002	0.028	0.004	NS
Suisun Slough at Rush Ranch	0.070	0.005	0.074	0.007	NS
Sacramento River at Hood DWR Station[3]	0.062	0.006	0.051	0.012	NS
Napa River at River Park Blvd.[4]	0.053	0.001	0.040	0.003	S** (75%)
Rough and Ready DWR station, Stockton	0.058	0.009	0.062	0.004	NS
Napa River at Vallejo Seawall (340)[5]	0.038	0.006	0.028	0.003	NS
Montezuma Slough at Nurse Slough (609)	0.068	0.006	0.064	0.005	NS
Suisun Bay off Chipps Island (508)	0.066	0.003	0.063	0.004	NS
Grizzly Bay at Dolphin (602)	0.066	0.011	0.065	0.006	NS
Carquinez Strait, West of Benicia army dock (405)[4]	0.048	0.005	0.057	0.003	NS
Trip Blank: DIEPAMHR	0.040	0.004	-	-	NA
Bottle Blank: DIEPAMHR	0.038	0.007	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.
*: $P < 0.05$
**: $P < 0.01$
2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.
3. This low conductivity sample was compared to the Low EC Control.
4. These high conductivity samples were compared to the High EC Control @ 14.06 mS/cm.
5. This high conductivity sample was compared to the High EC Control @ 23.81 mS/cm.

Table B 85-2. Summary of 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 6/9/09 - 6/10/09.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Suisun Slough at Rush Ranch	5680	18.0	7.37	7.2	51.3	0.09	0.001
Sacramento River at Hood DWR Station	171	21.3	7.48	8.3	16.4	0.33	0.004
Napa River at River Park Blvd.	13480	21.0	7.85	8.8	18.7	0.03	0.001
Rough and Ready DWR station, Stockton	552	23.3	7.51	6.2	12.7	0.07	0.001
Napa River at Vallejo Seawall (340)	23140	18.1	7.78	8.8	21.3	0.10	0.001
Montezuma Slough at Nurse Slough (609)	4481	19.6	7.70	8.5	63.5	0.12	0.002
Suisun Bay off Chipps Island (508)	2506	19.0	7.85	9.2	30.4	0.12	0.003
Grizzly Bay at Dolphin (602)	7520	18.7	8.00	9.3	129.3	0.13	0.003
Carquinez Strait, West of Benicia army dock (405)	12010	18.1	7.84	9.2	105.7	0.17	0.003
Trip Blank: DIEPAMHR	-	-	-	-	0.5	0.03	-
Bottle Blank: DIEPAMHR	-	-	-	-	0.3	0.04	-

Table B 85-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 6/11/09 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 6/9/09 - 6/10/09.

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	23.0	23.7	7.5	8.4	7.81	8.24	88	60	-
Low EC Control @ 161.5 µS/cm	157	23.0	23.4	7.6	8.6	7.43	8.27	48	22	-
High EC Control @ 14.06 mS/cm	13325	23.0	23.9	7.4	8.2	7.67	8.00	1640	96	-
High EC Control @ 23.81 mS/cm	22670	23.0	23.5	7.0	7.9	7.63	8.00	2800	88	-
Suisun Slough at Rush Ranch	5300	23.0	23.8	7.4	8.7	8.04	8.25	620	160	0.004
Sacramento River at Hood DWR Station	163	23.1	23.5	7.1	8.7	7.67	8.16	60	68	0.021
Napa River at River Park Blvd.	12740	23.1	24.0	7.0	8.3	7.94	8.07	1560	136	0.001
Rough and Ready DWR station, Stockton	545	23.0	23.7	7.2	8.7	7.82	8.20	148	76	0.005
Napa River at Vallejo Seawall (340)	22180	23.0	24.2	6.5	8.2	7.67	7.90	2760	108	0.002
Montezuma Slough at Nurse Slough (609)	4412	23.0	23.9	7.4	8.5	7.62	8.10	480	84	0.006
Suisun Bay off Chipps Island (508)	2269	23.1	24.2	7.6	8.4	7.80	8.03	272	74	0.005
Grizzly Bay at Dolphin (602)	7445	23.1	23.8	7.4	8.9	7.70	7.98	920	76	0.005
Carquinez Strait, West of Benicia army dock (405)	12135	23.1	24.1	7.0	8.2	7.69	7.90	1360	80	0.005
Trip Blank: DIEPAMHR	351	23.1	23.5	7.4	8.7	7.85	8.24	104	62	0.002
Bottle Blank: DIEPAMHR	338	23.1	24.2	7.5	8.6	7.80	8.17	104	64	0.003
DIEPAMHR + 25 ppb PBO	338	23.2	23.9	7.4	8.3	7.82	8.22	-	-	-
Low EC Control @ 161.5 µS/cm + 25 ppb PBO	160	23.2	24.0	7.5	8.4	7.46	8.16	-	-	-
High EC Control @ 14.06 mS/cm + 25 ppb PBO	13375	23.2	23.9	7.3	8.1	7.70	7.98	-	-	-
High EC Control @ 23.81 mS/cm + 25 ppb PBO	22580	23.2	24.1	7.1	8.3	7.74	8.02	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	5380	23.2	24.0	7.4	8.2	8.09	8.29	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	163.85	23.3	24.2	7.1	8.9	7.65	8.09	-	-	-
Napa River at River Park Blvd. + 25 ppb PBO	12835	23.3	24.0	7.0	8.2	7.93	8.03	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	531.5	23.4	24.2	7.3	8.5	7.83	8.04	-	-	-
Napa River at Vallejo Seawall (340) + 25 ppb PBO	22175	23.3	24.1	6.7	8.3	7.70	7.83	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	4334.5	23.4	24.1	7.4	8.5	7.86	8.02	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	2249.5	23.3	24.2	7.4	8.2	7.77	8.09	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	7420	23.4	23.9	7.5	8.2	7.60	7.97	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	12080	23.4	24.1	7.2	8.4	7.72	7.96	-	-	-

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 B 86-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 6/12/09 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 6/11/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	87	3.0	95	2.9	S* (109%)
Low EC Control @ 168.2 µS/cm	90	4.1	84*	3.2	NS
Sacramento R. Deep Water Channel, Light 55	95	5.0	89	4.1	NS
Upper Cache Slough at mouth of Ulatis Creek[3]	92	5.3	82	2.6	NS
Confluence of Linsey Sl. And Cache Sl.[3]	81	11.2	95	3.1	NS
Sacramento River at tip of Grand Island (711)[3]	78	5.7	72	8.4	NS
San Joaquin River at Potato Slough (815)[3]	93	7.5	93	2.5	NS
Old River, western arm at railroad bridge (902)[3]	98	2.5	84	5.2	NS
Old River at mouth of Holland Cut (915)	92	5.3	90	7.1	NS
Field Dup.: San Joaquin River at Potato Slough (815)[3]	92	5.3	-	-	NA
Bottle Blank: DIEPAMHR	84	2.6	-	-	NA
Bottle Blank: Clear Plastic	86	5.9	-	-	NA
Bottle Blank: Amber Plastic	95	5.0	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.027	0.009	0.037	0.003	NS
Low EC Control @ 168.2 µS/cm	0.029	0.008	0.042	0.007	NS
Sacramento R. Deep Water Channel, Light 55	0.086	0.006	0.064	0.002	S* (74%)
Upper Cache Slough at mouth of Ulatis Creek[3]	0.075	0.014	0.084	0.012	NS
Confluence of Linsey Sl. And Cache Sl.[3]	0.067	0.003	0.068	0.006	NS
Sacramento River at tip of Grand Island (711)[3]	0.056	0.006	0.073	0.009	NS
San Joaquin River at Potato Slough (815)[3]	0.079	0.005	0.091	0.009	NS
Old River, western arm at railroad bridge (902)[3]	0.081	0.004	0.060	0.009	S* (74%)
Old River at mouth of Holland Cut (915)	0.070	0.003	0.078	0.004	NS
Field Dup.: San Joaquin River at Potato Slough (815)[3]	0.045	0.009	-	-	NA
Bottle Blank: DIEPAMHR	0.048	0.006	-	-	NA
Bottle Blank: Clear Plastic	0.043	0.007	-	-	NA
Bottle Blank: Amber Plastic	0.060	0.003	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard statistical protocols.

*: $P < 0.05$

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

3. These low conductivity samples were compared to the Low EC Control.

Table B 86-2. Summary of 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 6/11/09.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (^{o}C)	pH	DO (mg/L)			
Sacramento R. Deep Water Channel, Light 55	255	19.6	7.96	8.9	44.7	0.10	0.003
Upper Cache Slough at mouth of Ulatis Creek	214	18.9	7.96	9.0	101.9	0.08	0.002
Confluence of Lindsey Sl. And Cache Sl.	183	19.2	7.86	8.8	51.3	0.07	0.002
Sacramento River at tip of Grand Island (711)	170	20.2	7.66	8.6	11.4	0.10	0.002
San Joaquin River at Potato Slough (815)	182	20.7	7.87	8.9	6.7	0.00	0.000
Old River, western arm at railroad bridge (902)	213	21.7	7.80	8.5	6.3	0.00	0.000
Old River at mouth of Holland Cut (915)	271	22.1	7.80	8.1	5.2	0.00	0.000
Field Dup.: San Joaquin River at Potato Slough (815)	182	20.7	7.87	8.9	6.3	0.00	0.000
Bottle Blank: DIEPAMHR	-	-	-	-	0.5	0.00	-
Bottle Blank: Clear Plastic	-	-	-	-	-	-	-
Bottle Blank: Amber Plastic	-	-	-	-	-	-	-

Table B 86-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 6/12/09 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 6/11/09.

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 (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	337	22.8	24.3	7.3	8.1	7.72	8.11	88	60	-
Low EC Control @ 168.2 µS/cm	172	22.8	24.2	7.2	8.4	7.54	7.97	52	30	-
Sacramento R. Deep Water Channel, Light 55	248	22.8	24.4	7.2	8.3	7.64	8.10	76	72	0.006
Upper Cache Slough at mouth of Ulatis Creek	199	22.8	24.3	7.4	8.4	7.78	8.22	68	70	0.006
Confluence of Linsey Sl. And Cache Sl.	180	22.9	24.3	7.2	8.4	7.71	8.08	64	64	0.004
Sacramento River at tip of Grand Island (711)	163	23.0	24.4	7.1	8.3	7.68	8.03	56	64	0.005
San Joaquin River at Potato Slough (815)	178	22.9	24.4	7.1	8.4	7.63	8.09	64	56	0.000
Old River, western arm at railroad bridge (902)	210	22.9	24.4	7.3	8.2	7.71	8.12	60	60	0.000
Old River at mouth of Holland Cut (915)	261	23.0	24.3	7.4	8.4	7.74	8.06	72	62	0.000
Field Dup.: San Joaquin River at Potato Slough (815)	181	23.1	24.4	7.1	8.7	7.69	7.96	60	58	0.000
Bottle Blank: DIEPAMHR	340	23.1	24.3	7.3	8.3	7.76	8.11	104	58	0.000
Bottle Blank: Clear Plastic	339	23.2	24.3	7.5	8.4	7.76	8.10	88	60	-
Bottle Blank: Amber Plastic	342	23.2	24.4	7.4	8.8	7.76	8.11	88	60	-
DIEPAMHR + 25 ppb PBO	339	23.2	23.7	7.2	8.2	7.78	8.19	-	-	-
Low EC Control @ 168.2 µS/cm + 25 ppb PBO	172	23.3	23.6	7.3	8.5	7.48	7.94	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	246	23.3	23.9	7.4	8.2	7.78	8.13	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	201	23.3	23.9	7.6	8.7	7.79	8.20	-	-	-
Confluence of Linsey Sl. And Cache Sl. + 25 ppb PBO	180	23.5	23.7	7.3	8.3	7.73	8.11	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	163	23.4	23.9	7.3	8.6	7.66	8.06	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	179	23.5	23.9	7.3	8.4	7.71	8.08	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	208	23.6	23.9	7.3	8.4	7.74	8.02	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	264	23.6	23.8	7.3	8.4	7.78	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 B 87-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 6/25/09 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 6/23/09 - 6/24/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	92	2.7	95	2.9	NS
Low EC Control @ 140.9 µS/cm	95	2.6	91	5.1	NS
High EC Control @ 12.53 mS/cm	100	0.0	95	5.0	NS
High EC Control @ 17.69 mS/cm	98	2.5	93	4.4	NS
High EC Control @ 20.23 mS/cm	80	12.2	79	4.1	NS
Napa River, near River Park Blvd. [5]	92	5.3	93	7.5	NS
Suisun Slough @ Rush Ranch	98	2.5	97	2.8	NS
Rough and Ready DWR Station, Stockton	97	2.8	100	0.0	NS
Sacramento River at Hood DWR Station [3]	87*	3.0	66**	6.1	S* (76%)
Carquinez Strait, West of Benicia army dock (405) [6]	87	10.2	90	6.7	NS
Montezuma Slough at Nurse Slough (609)	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) [4]	97	2.8	100	0.0	NS
Trip Blank (DIEPAMHR)	97	2.8	-	-	NA
Trip Blank (DIEPAMHR)	98	2.5	-	-	NA
Field Dup: Carquinez Strait, West of Benicia army dock (405) [6]	91	5.4	-	-	NA
Field Dup: Suisun Bay off Chipps Island (508)	100	0.0	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.041	0.012	0.049	0.004	NS
Low EC Control @ 140.9 µS/cm	0.041	0.004	0.042	0.005	NS
High EC Control @ 12.53 mS/cm	0.044	0.006	0.048	0.006	NS
High EC Control @ 17.69 mS/cm	0.037	0.004	0.042	0.005	NS
High EC Control @ 20.23 mS/cm	0.037	0.001	0.028	0.006	NS
Napa River, near River Park Blvd. [5]	0.043	0.005	0.042	0.005	NS
Suisun Slough @ Rush Ranch	0.089	0.011	0.093	0.017	NS
Rough and Ready DWR Station, Stockton	0.075	0.005	0.133	0.029	NS
Sacramento River at Hood DWR Station [3]	0.035	0.006	0.068	0.015	NS
Carquinez Strait, West of Benicia army dock (405) [6]	0.043	0.012	0.026	0.007	NS
Montezuma Slough at Nurse Slough (609)	0.063	0.005	0.060	0.007	NS
Suisun Bay off Chipps Island (508)	0.052	0.009	0.047	0.010	NS
Grizzly Bay at Dolphin (602) [4]	0.068	0.004	0.060	0.004	NS
Trip Blank (DIEPAMHR)	0.061	0.005	-	-	NA
Trip Blank (DIEPAMHR)	0.046	0.005	-	-	NA
Field Dup: Carquinez Strait, West of Benicia army dock (405) [6]	0.057	0.006	-	-	NA
Field Dup: Suisun Bay off Chipps Island (508)	0.061	0.012	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.
*: $P < 0.05$
**: $P < 0.01$
2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.
3. This low conductivity sample was compared to the Low EC Control.
4. This high conductivity sample was compared to the High EC Control @ 12.53 mS/cm.
5. This high conductivity sample was compared to the High EC Control @ 17.69 mS/cm.
6. These high conductivity samples were compared to the High EC Control @ 20.23 mS/cm.

Table B 87-2. Summary of 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 6/23/09 - 6/24/09.

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, Near River Park Blvd.	17150	22.3	7.51	6.9	14.0	0.04	0.000
Suisun Slough at Rush Ranch	6460	20.4	7.13	5.5	51.1	0.15	0.001
Rough and Ready DWR Station, Stockton	607	24.2	7.34	6.0	11.5	0.09	0.001
Sacramento River at Hood DWR Station	149	23.2	7.11	7.5	20.4	0.30	0.002
Carquinez Strait, west of Benicia army dock (405)	19430	19.7	7.45	8.9	240.7	0.20	0.001
Montezuma Slough at Nurse Slough (609)	5750	22.0	7.46	7.5	73.6	0.11	0.001
Suisun Bay, off Chipps Island (508)	8510	20.5	7.60	8.8	24.6	0.08	0.001
Grizzly Bay at Dolphin (602)	11900	21.1	7.75	8.9	177.3	0.17	0.004
Trip Blank (DIEPAMHR) 6/23/09	-	-	-	-	0.4	0.02	-
Trip Blank (DIEPAMHR) 6/24/09	-	-	-	-	0.3	0.00	-
Field Dup: Carquinez Strait, west of Benicia army dock (405)	19430	19.7	7.45	8.9	276.3	0.27	0.002
Field Dup: Suisun Bay, off Chipps Island (508)	8510	20.5	7.60	8.8	24.4	0.10	0.001

Table B 87-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 6/25/09 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 6/23/09 - 6/24/09.

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	280	22.7	23.2	7.1	8.4	6.90	8.13	100	58	-
Low EC Control @ 140.9 µS	149	22.8	23.1	7.4	8.7	6.98	7.84	40	26	-
High EC Control @ 12.53 mS	11240	22.9	23.3	7.3	8.6	6.90	7.91	1440	74	-
High EC Control @ 17.69 mS	16970	22.9	23.6	7.1	8.6	6.99	8.00	1960	84	-
High EC Control @ 20.23 mS	19720	22.8	23.4	7.2	8.3	7.20	7.99	2360	84	-
Napa River, near River Park Blvd.	16855	22.9	23.8	7.0	8.5	7.42	7.75	2280	134	0.001
Suisun Slough @ Rush Ranch	6265	22.9	23.9	7.0	8.5	7.56	7.97	710	138	0.004
Rough and Ready DWR Station, Stockton	626	22.9	23.7	7.3	8.9	7.50	8.00	130	82	0.004
Sacramento River at Hood DWR Station	148	22.9	23.7	6.9	8.9	7.07	7.67	48	54	0.007
Carquinez Strait, West of Benicia army dock (405)	18475	22.9	23.6	6.6	8.0	7.04	7.78	2160	86	0.004
Montezuma Slough at Nurse Slough (609)	5585	22.6	23.6	7.4	8.8	7.39	7.78	620	92	0.003
Suisun Bay off Chipps Island (508)	7740	22.9	23.5	5.9	8.5	7.15	7.81	840	70	0.002
Grizzly Bay at Dolphin (602)	11470	22.8	23.5	7.0	8.9	7.26	7.85	1320	76	0.004
Trip Blank (DIEPAMHR)	417	22.9	23.7	7.5	8.7	7.28	8.25	104	58	0.002
Trip Blank (DIEPAMHR)	390	22.9	24.1	7.1	8.6	7.37	8.25	100	58	0.000
Field Dup: Carquinez Strait, West of Benicia army dock (405)	18880	22.9	24.3	7.0	8.4	7.25	7.75	2160	84	0.005
Field Dup: Suisun Bay off Chipps Island (508)	8180	23.0	23.4	7.2	8.7	7.25	7.77	840	70	0.002
DIEPAMHR + 25 ppb PBO	410	22.8	23.0	7.3	8.3	7.13	8.13	-	-	-
Low EC Control @ 140.9 µS + 25 ppb PBO	161	22.5	23.1	7.1	8.6	7.13	7.92	-	-	-
High EC Control @ 12.53 mS + 25 ppb PBO	12060	23.1	23.2	6.9	8.5	6.94	7.97	-	-	-
High EC Control @ 17.69 mS + 25 ppb PBO	16970	23.0	23.1	6.9	8.7	7.23	7.97	-	-	-
High EC Control @ 20.23 mS + 25 ppb PBO	19670	23.2	23.3	7.0	8.4	7.29	8.00	-	-	-
Napa River, near River Park Blvd. + 25 ppb PBO	16855	23.2	23.2	6.8	8.3	7.37	7.83	-	-	-
Suisun Slough @ Rush Ranch + 25 ppb PBO	6395	23.2	23.3	7.0	8.8	7.46	7.96	-	-	-
Rough and Ready DWR Station, Stockton + 25 ppb PBO	630	23.0	23.2	7.3	8.5	7.26	8.14	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	151	23.2	23.6	7.0	8.5	7.15	7.89	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	18965	23.2	23.8	6.6	8.0	7.11	7.77	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	5640	23.3	23.3	7.4	8.5	7.32	7.81	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	8000	23.2	23.8	7.1	8.8	7.18	8.83	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	11490	23.3	23.4	7.0	8.5	7.30	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 B 88-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 6/26/09 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 6/25/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	97	3.1	95	2.8	NS
Low EC Control @ 132.6 µS/cm	89	6.4	77	6.1	NS
Sacramento River Deep Water Channel, Light 55	87	3.0	88	4.8	NS
Upper Cache Slough at mouth of Ulatis Creek	84	7.1	74	11.6	NS
Confluence of Lindsey Sl. and Cache Sl.	74	15.4	89	0.6	NS
Sacramento River at tip of Grand Island (711) [3]	45**	7.6	61*	4.2	NS
San Joaquin River at Potato Slough (815)	85	11.9	84	9.7	NS
Old River, western arm at railroad bridge (902)	90	7.1	85*	4.2	NS
Old River at mouth of Holland Cut (915)	85	6.4	88	7.5	NS
Bottle Blank Clear (cubitainer)	78	7.9	-	-	NS
Bottle Blank Amber (cubitainer)	93	2.5	-	-	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.046	0.010	0.044	0.005	NS
Low EC Control @ 132.6 µS/cm	0.042	0.007	0.035	0.003	NS
Sacramento River Deep Water Channel, Light 55	0.072	0.011	0.057	0.002	NS
Upper Cache Slough at mouth of Ulatis Creek	0.085	0.005	0.072	0.004	NS
Confluence of Lindsey Sl. and Cache Sl.	0.083	0.007	0.043	0.007	S** (52%)
Sacramento River at tip of Grand Island (711) [3]	0.075	0.013	0.054	0.011	NS
San Joaquin River at Potato Slough (815)	0.067	0.009	0.058	0.006	NS
Old River, western arm at railroad bridge (902)	0.061	0.009	0.077	0.009	NS
Old River at mouth of Holland Cut (915)	0.055	0.010	0.078	0.007	S* (142%)
Bottle Blank Clear (cubitainer)	0.038	0.010	-	-	NS
Bottle Blank Amber (cubitainer)	0.026	0.003	-	-	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

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

3. This low conductivity sample was compared to the Low EC Control @ 312.6 uS.

Table B 88-2. 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 6/25/09.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (^{o}C)	pH	DO (mg/L)			
Sacramento River, Deep Water Channel, Light 55	246	22.0	7.69	8.5	29.7	0.05	0.001
Upper Cache Slough at Mouth of Ulatis Creek	207	20.9	7.62	8.8	60.8	0.04	0.001
Confluence of Lindsey Sl. and Cache Sl.	188	22.1	7.50	8.6	27.9	0.10	0.001
Sacramento River at tip of Grand Island (711)	134	23.2	7.37	8.1	10.6	0.19	0.002
San Joaquin River at Potato Slough (815)	182	21.9	7.47	8.5	6.3	0.03	0.000
Old River, western arm at railroad bridge (902)	204	22.4	7.90	8.5	5.3	0.03	0.001
Old River at mouth of Holland Cut (915)	217	23.3	7.63	7.9	4.3	0.01	0.000

Table B 88-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 6/26/09 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 6/25/09.

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	334	22.5	23.1	6.8	8.3	7.52	8.02	-	-	-
Low EC Control @ 132.6 µS/cm	128	22.5	23.4	7.3	8.4	7.26	7.79	40	26	-
Sacramento River Deep Water Channel, Light 55	232	22.6	23.2	6.5	8.5	7.57	8.04	76	74	0.002
Upper Cache Slough at mouth of Ulatis Creek	196	22.3	23.3	7.0	8.6	7.43	8.08	68	74	0.002
Confluence of Lindsey Sl. and Cache Sl.	174	22.5	23.2	6.7	8.6	7.42	7.97	72	66	0.004
Sacramento River at tip of Grand Island (711)	127	22.3	23.3	6.9	8.4	7.39	7.80	60	52	0.006
San Joaquin River at Potato Slough (815)	171	22.3	22.9	6.9	8.7	7.41	8.02	64	60	0.001
Old River, western arm at railroad bridge (902)	194	22.5	23.4	6.8	8.5	7.42	8.00	60	58	0.001
Old River at mouth of Holland Cut (915)	217	22.8	23.3	7.0	8.4	7.53	7.93	72	58	0.000
Bottle Blank Clear (cubitainer)	324	22.4	23.1	6.9	8.5	7.44	8.06	-	-	-
Bottle Blank Amber (cubitainer)	325	22.8	23.2	7.1	8.6	7.39	8.08	-	-	-
DIEPAMHR + 25 ppb PBO	325	22.5	23.3	7.1	8.3	7.59	8.06	-	-	-
Low EC Control @ 132.6 µS + 25 ppb PBO	134	22.2	23.6	7.1	8.5	7.23	7.75	-	-	-
Sacramento River Deep Water Channel, Light 55 + 25 ppb PBO	234	22.7	23.4	6.4	8.6	7.55	8.00	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	191	22.3	22.4	6.9	8.5	7.40	8.02	-	-	-
Confluence of Lindsey Sl. and Cache Sl. + 25 ppb PBO	172	22.4	23.5	6.9	8.5	7.46	7.99	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	131	22.4	23.5	7.2	8.7	7.41	7.89	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	180	22.5	23.3	6.6	8.7	7.33	8.07	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	212	22.8	23.6	7.0	8.5	7.38	7.95	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	194	22.3	23.3	6.8	8.8	7.53	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 B 89-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 7/09/09 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 7/07/09-7/08/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	98	2.5	98	2.5	NS
Low EC Control @ 122.9 µS/cm	98	2.5	98	2.5	NS
High EC Control @ 11.68 mS/cm	100	0.0	100	0.0	NS
High EC Control @ 21.28 mS/cm	89	7.9	93	2.5	NS
Napa River, near River Park Blvd. [5]	98	2.5	97	3.1	NS
Sacramento River at Hood DWR Station [3]	95	2.9	98	2.5	NS
Rough and Ready DWR Station, Stockton	98	2.5	98	2.5	NS
Suisun Slough @ Rush Ranch	100	0.0	100	0.0	NS
Suisun Bay off Chipps Island (508)	100	0.0	97	2.8	NS
Montezuma Slough at Nurse Slough (609)	100	0.0	100	0.0	NS
Grizzly Bay at Dolphin (602) [4]	95	2.9	100	0.0	NS
Carquinez Strait, West of Benicia army dock (405) [5]	79	7.2	87	5.1	NS
Field Dup: Napa River, near River Park Blvd. [5]	92	8.3	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.067	0.004	0.077	0.004	NS
Low EC Control @ 122.9 µS/cm	0.057	0.006	0.058*	0.004	NS
High EC Control @ 11.68 mS/cm	0.051*	0.005	0.057**	0.001	NS
High EC Control @ 21.28 mS/cm	0.031***	0.001	0.033***	0.005	NS
Napa River, near River Park Blvd. [5]	0.039	0.004	0.012*	0.002	S* (31%)
Sacramento River at Hood DWR Station [3]	0.094	0.005	0.056	0.006	S** (60%)
Rough and Ready DWR Station, Stockton	0.109	0.010	0.089	0.009	NS
Suisun Slough @ Rush Ranch	0.109	0.010	0.104	0.008	NS
Suisun Bay off Chipps Island (508)	0.097	0.004	0.109	0.015	NS
Montezuma Slough at Nurse Slough (609)	0.102	0.006	0.097	0.006	NS
Grizzly Bay at Dolphin (602) [4]	0.079	0.004	0.081	0.004	NS
Carquinez Strait, West of Benicia army dock (405) [5]	0.069	0.005	0.061	0.003	NS
Field Dup: Napa River, near River Park Blvd. [5]	0.044	0.001	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

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

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

4. This high conductivity sample was compared to the High EC Control @ 11.68 mS/cm.

5. These high conductivity samples were compared to the High EC Control @ 21.28 mS/cm.

Table B 89-2. Summary of 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 7/7/09 - 7/8/09.

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, near River Park Blvd.	20510	21.3	7.47	7.6	11.6	0.16	0.001
Sacramento River at Hood DWR Station	146	21.6	6.93	8.4	17.2	0.19	0.001
Rough and Ready DWR Station, Stockton	584	25.2	7.15	7.8	7.7	0.07	0.001
Suisun Slough @ Rush Ranch	7520	17.3	7.07	7.5	55.0	0.19	0.001
Suisun Bay off Chipps Island (508)	6050	19.7	7.49	9.0	19.5	0.07	0.001
Montezuma Slough at Nurse Slough (609)	6470	20.8	7.44	8.2	38.7	0.06	0.001
Grizzly Bay at Dolphin (602)	13140	19.6	7.62	9.1	87.0	0.13	0.001
Carquinez Strait, West of Benicia army dock (405)	19350	18.9	7.41	9.0	124.7	0.18	0.001
Field Dup: Napa River, near River Park Blvd.	20510	21.3	7.47	7.6	11.9	0.04	0.000

Table B 89-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 7/9/09 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 7/7/09 - 7/8/09.

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 (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	351	23.9	24.1	7.0	8.6	7.68	8.21	100	58	-
Low EC Control @ 122.9 µS/cm	140	23.8	23.9	7.1	8.7	7.32	7.85	40	28	-
High EC Control @ 11.68 mS/cm	11265	24.0	24.0	6.8	8.3	7.54	7.92	1320	76	-
High EC Control @ 21.28 mS/cm	21085	24.0	24.2	6.0	8.6	7.47	7.94	2520	84	-
Napa River, near River Park Blvd.	19620	23.8	24.2	6.2	8.4	7.64	7.83	2480	134	0.002
Sacramento River at Hood DWR Station	136	23.8	24.2	6.4	8.8	7.44	7.72	64	50	0.005
Rough and Ready DWR Station, Stockton	571	23.7	24.2	7.0	8.5	7.78	7.90	130	82	0.002
Suisun Slough @ Rush Ranch	7090	23.9	24.2	7.0	8.3	7.73	8.06	920	164	0.004
Suisun Bay off Chipps Island (508)	5815	23.9	24.2	6.9	8.7	6.78	7.82	640	74	0.002
Montezuma Slough at Nurse Slough (609)	6070	23.9	24.2	6.8	8.4	7.73	7.91	700	94	0.002
Grizzly Bay at Dolphin (602)	12605	23.9	24.2	6.9	8.3	7.60	7.86	1480	86	0.003
Carquinez Strait, West of Benicia army dock (405)	18300	23.8	24.4	6.6	8.3	7.60	7.77	2200	86	0.004
Field Dup: Napa River, near River Park Blvd.	19760	23.7	24.3	6.6	8.8	7.58	7.85	2440	134	0.001
DIEPAMHR	340	22.9	24.3	7.2	8.6	7.70	8.00	-	-	-
Low EC Control @ 122.9 µS/cm	155	23.0	24.3	7.1	8.6	7.33	7.82	-	-	-
High EC Control @ 11.68 mS/cm	11130	23.0	24.3	6.9	8.6	7.52	7.74	-	-	-
High EC Control @ 21.28 mS/cm	20815	23.4	24.5	6.2	8.0	7.52	7.73	-	-	-
Napa River, near River Park Blvd.	19455	23.6	24.2	6.4	8.2	7.56	7.81	-	-	-
Sacramento River at Hood DWR Station	646	22.8	24.3	6.5	8.7	7.38	7.82	-	-	-
Rough and Ready DWR Station, Stockton	584	23.6	24.3	6.9	8.4	7.76	7.90	-	-	-
Suisun Slough @ Rush Ranch	7156	23.4	24.2	6.4	8.3	7.73	8.05	-	-	-
Suisun Bay off Chipps Island (508)	5700	22.6	24.5	7.1	8.6	7.56	7.80	-	-	-
Montezuma Slough at Nurse Slough (609)	6020	22.5	24.3	7.0	8.5	7.72	7.83	-	-	-
Grizzly Bay at Dolphin (602)	12615	23.5	24.5	7.0	8.3	7.67	7.75	-	-	-
Carquinez Strait, West of Benicia army dock (405)	18285	22.6	24.3	6.6	8.4	7.61	7.70	-	-	-

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 B 90-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 7/10/09 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 7/09/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	98	2.5	98	2.5	NA
Low EC Control @ 167.1 µS/cm	100	0.0	98	2.5	NA
Sacramento R. Deep Water Channel, Light 55	100	0.0	100	0.0	NA
Upper Cache Slough at mouth of Ulatis Creek[3]	100	0.0	100	0.0	NA
Confluence of Lindsey Sl. And Cache Sl.[3]	95	4.5	90	3.5	NA
Sacramento River at tip of Grand Island (711)[3]	93	3.3	83*	2.2	NA
San Joaquin River at Potato Slough (815)	98	2.5	98	2.3	NA
Old River, western arm at railroad bridge (902)	98	2.5	100	0.0	NA
Old River at mouth of Holland Cut (915)	100	0.0	100	0.0	NA
Trip Blank: DIEPAMHR	95	2.9	-	-	NA
Field Dup.: Confluence of Lindsey Sl. And Cache Sl.[3]	92	4.8	-	-	NA
Bottle Blank: DIEPAMHR	92	5.3	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.068	0.004	0.051	0.008	S* (75%)
Low EC Control @ 167.1 µS/cm	0.063	0.009	0.027*	0.003	S** (40%)
Sacramento R. Deep Water Channel, Light 55	0.106	0.010	0.087	0.007	NA
Upper Cache Slough at mouth of Ulatis Creek[3]	0.075	0.006	0.064	0.005	NA
Confluence of Lindsey Sl. And Cache Sl.[3]	0.083	0.003	0.086	0.006	NA
Sacramento River at tip of Grand Island (711)[3]	0.032	0.014	0.055	0.007	NA
San Joaquin River at Potato Slough (815)	0.064	0.006	0.069	0.002	NA
Old River, western arm at railroad bridge (902)	0.084	0.014	0.089	0.003	NA
Old River at mouth of Holland Cut (915)	0.112	0.005	0.081	0.010	S* (72%)
Trip Blank: DIEPAMHR	0.067	0.005	-	-	NA
Field Dup.: Confluence of Lindsey Sl. And Cache Sl.[3]	0.094	0.009	-	-	NA
Bottle Blank: DIEPAMHR	0.057	0.007	-	-	NA

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

*: $P < 0.05$

**: $P < 0.01$

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

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

Table B 90-2. Summary of 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 7/09/09.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (^{o}C)	pH	DO (mg/L)			
Sacramento R. Deep Water Channel, Light 55	244	21.0	8.14	8.5	37.1	0.09	0.005
Upper Cache Slough at mouth of Ulatis Creek	186	19.6	7.79	8.7	77.1	0.10	0.002
Confluence of Lindsey Sl. And Cache Sl.	155	20.4	7.66	8.6	41.9	0.19	0.003
Sacramento River at tip of Grand Island (711)	117	22.1	7.36	8.3	20.3	0.22	0.002
San Joaquin River at Potato Slough (815)	160	22.6	7.49	8.2	6.3	0.21	0.003
Old River, western arm at railroad bridge (902)	253	23.1	7.66	8.3	4.6	0.09	0.002
Old River at mouth of Holland Cut (915)	241	23.2	7.66	8.1	4.8	0.04	0.001
Trip Blank	-	-	-	-	0.4	0.03	-
Field Dup.: Confluence of Lindsey Sl. And Cache Sl.	155	20.4	7.66	8.6	44.3	0.20	0.003
Bottle Blank	-	-	-	-	0.3	0.02	-

Table B 90-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 7/10/09 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 7/09/09.

Treatment	Laboratory Chemistry								Hardness (mg/L as $CaCO_3$)	Alkalinity (mg/L as $CaCO_3$)	Unionized Ammonia (mg/L)[1]
	SC (uS/cm)	EC (uS/cm)	Min Temp (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	325	316	23.7	23.7	7.0	8.5	7.65	8.10	100	58	-
Low EC Control @ 167.1 µS/cm	176	171	23.6	23.8	7.1	8.6	7.40	7.92	56	30	-
Sacramento R. Deep Water Channel, Light 55	249	242	23.6	23.8	7.1	8.5	7.71	8.06	92	74	0.005
Upper Cache Slough at mouth of Ulatis Creek	189	184	23.6	23.7	7.0	8.8	7.72	8.02	76	70	0.005
Confluence of Lindsey Sl. And Cache Sl.	157	153	23.5	23.7	7.2	8.5	7.59	7.97	56	58	0.008
Sacramento River at tip of Grand Island (711)	121	117	23.6	23.7	6.9	8.9	7.54	7.89	48	48	0.008
San Joaquin River at Potato Slough (815)	216	210	23.6	23.6	7.0	8.9	7.59	7.87	48	52	0.007
Old River, western arm at railroad bridge (902)	254	245	22.7	23.6	7.0	8.8	7.68	8.11	76	58	0.005
Old River at mouth of Holland Cut (915)	243	237	23.5	23.6	6.9	8.5	7.58	8.11	64	60	0.002
Trip Blank	346	336	23.6	23.8	7.3	8.5	7.66	8.07	100	60	0.002
Field Dup.: Confluence of Lindsey Sl. And Cache Sl.	156	152	23.3	23.5	7.2	8.8	7.66	7.97	68	58	0.009
Bottle Blank	346	339	23.5	24.7	7.3	8.2	7.69	8.07	100	58	0.001
DIEPAMHR + 25 ppb PBO	322	310	23.1	23.6	7.2	8.2	7.65	8.06	-	-	-
Low EC Control @ 167.1 µS/cm + 25 ppb PBO	175	170	23.6	23.6	7.3	8.6	6.70	7.87	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	248	241	23.4	23.6	7.1	8.5	7.72	8.05	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	187	184	23.7	23.9	7.2	8.4	7.74	8.12	-	-	-
Confluence of Lindsey Sl. And Cache Sl. + 25 ppb PBO	157	154	23.8	24.2	7.1	8.7	7.62	7.96	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	120	117	23.8	24.1	7.0	8.4	7.55	7.91	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	163	160	23.5	24.9	6.9	8.6	7.58	7.92	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	258	254	23.9	24.4	7.1	8.8	7.65	8.05	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	242	238	23.6	24.8	6.9	8.7	7.62	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 B 91-1. Summary of 10-day H. azteca water column toxicity test initiated on 7/22/09 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 7/207/09-7/21/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	100	0.0	98	2.5	NS
Low EC Control @ 112.0 µS/cm	100	0.0	100	0.0	NS
High EC Control @ 18.56 mS/cm	97	2.8	100	0.0	NS
High EC Control @ 23.47 mS/cm	93	7.5	95	2.9	NS
Suisun Bay off Chipps Island (508)	98	2.5	95	2.9	NS
Montezuma Slough at Nurse Slough (609)	98	2.3	98	2.5	NS
Grizzly Bay at Dolphin (602) [4]	93	4.8	82*	4.3	NS
Carquinez Strait, West of Benecia army dock (405) [5]	90	4.1	84	9.7	NS
Old River at mouth of Holland Cut (915)	92	2.7	100	0.0	NS
Sacramento River at tip of Grand Island (711) [3]	98	2.5	100	0.0	NS
Old River, western arm at railroad bridge (902)	100	0.0	100	0.0	NS
Confluence of Lindsey Sl. and Cache Sl.	100	0.0	100	0.0	NS
San Joaquin River at Potato Slough (815)	98	2.5	100	0.0	NS
Upper Cache Slough at mouth of Ulatis Creek	100	0.0	100	0.0	NS
Sacramento River Deep Water Channel, Light 55	98	2.5	98	2.5	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.072	0.008	0.063	0.008	NS
Low EC Control @ 112.0 µS/cm	0.052	0.011	0.054	0.004	NS
High EC Control @ 18.56 mS/cm	0.058	0.004	0.043*	0.005	S*
High EC Control @ 23.47 mS/cm	0.028	0.008	0.038*	0.003	NS
Suisun Bay off Chipps Island (508)	0.116	0.006	0.113	0.011	NS
Montezuma Slough at Nurse Slough (609)	0.116	0.006	0.116	0.007	NS
Grizzly Bay at Dolphin (602) [4]	0.046*	0.001	0.074	0.005	S***
Carquinez Strait, West of Benecia army dock (405) [5]	0.031	0.006	0.066	0.007	S**
Old River at mouth of Holland Cut (915)	0.120	0.009	0.109	0.007	NS
Sacramento River at tip of Grand Island (711) [3]	0.087	0.003	0.090	0.005	NS
Old River, western arm at railroad bridge (902)	0.124	0.003	0.107	0.008	NS
Confluence of Lindsey Sl. and Cache Sl.	0.105	0.011	0.092	0.005	NS
San Joaquin River at Potato Slough (815)	0.125	0.002	0.134	0.006	NS
Upper Cache Slough at mouth of Ulatis Creek	0.101	0.008	0.088	0.005	NS
Sacramento River Deep Water Channel, Light 55	0.128	0.005	0.117	0.003	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.
*: $P < 0.05$
**: $P < 0.01$
2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.

3. This low conductivity sample was compared to the Low EC Control.
4. This high conductivity sample was compared to the High EC Control @ 18.56 mS/cm.
5. This high conductivity sample was compared to the High EC Control @ 23.47 mS/cm.

Table B 91-2. Summary of 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 7/20/09 - 7/21/09.

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)	4457	21.7	7.43	8.5	16.0	0.07	0.001
Montezuma Slough at Nurse Slough (609)	6970	22.4	7.41	7.4	31.7	0.05	0.000
Grizzly Bay at Dolphin (602)	17770	20.3	7.73	8.6	215.7	0.19	0.003
Carquinez Strait, West of Benecia army dock (405)	22330	19.9	7.69	8.3	45.1	0.07	0.001
Old River at mouth of Holland Cut (915)	322	24.3	7.56	8.0	4.5	0.00	0.000
Sacramento River at tip of Grand Island (711)	111	21.3	7.26	8.4	22.1	0.08	0.001
Old River, western arm at railroad bridge (902)	386	23.9	7.65	8.4	4.2	0.00	0.000
Confluence of Lindsey Sl. and Cache Sl.	163	19.8	7.57	9.0	36.3	0.09	0.001
San Joaquin River at Potato Slough (815)	244	22.4	7.48	8.2	6.1	0.03	0.000
Upper Cache Slough at mouth of Ulatis Creek	136	20.2	7.32	8.8	68.2	0.04	0.000
Sacramento River Deep Water Channel, Light 55	249	21.6	7.62	8.3	24.4	0.04	0.001

Table B 91-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 7/22/09 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 7/20/09 - 7/21/09.

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 (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	345	23.3	24.3	7.1	8.1	7.63	7.95	100	58	-
Low EC Control @ 112.0 µS/cm	126	23.3	24.3	7.2	8.4	7.27	7.76	44	20	-
High EC Control @ 18.56 mS/cm	17540	23.3	24.3	7.0	8.2	7.53	7.97	2120	86	-
High EC Control @ 23.47 mS/cm	22445	23.8	24.3	6.9	8.6	7.52	7.95	2880	96	-
Suisun Bay off Chipps Island (508)	4198	23.3	24.3	7.2	8.6	7.52	7.95	464	64	0.003
Montezuma Slough at Nurse Slough (609)	6575	23.4	24.3	7.0	8.2	7.66	7.94	800	94	0.002
Grizzly Bay at Dolphin (602)	17060	23.3	24.3	6.8	8.3	7.59	7.86	2000	82	0.005
Carquinez Strait, West of Benecia army dock (405)	21390	23.3	24.0	6.6	8.8	7.53	7.84	2560	86	0.002
Old River at mouth of Holland Cut (915)	354	23.4	24.2	7.1	8.2	7.57	8.15	72	54	0.000
Sacramento River at tip of Grand Island (711)	125	23.4	24.2	6.6	8.1	7.43	8.05	48	48	0.004
Old River, western arm at railroad bridge (902)	392	23.3	24.0	7.3	8.4	7.54	8.10	80	54	0.000
Confluence of Lindsey Sl. and Cache Sl.	151	23.3	24.1	7.3	8.3	7.59	8.09	56	54	0.005
San Joaquin River at Potato Slough (815)	251	23.3	24.2	6.5	8.0	7.53	8.03	64	50	0.002
Upper Cache Slough at mouth of Ulatis Creek	180	23.2	24.2	7.3	8.2	7.69	8.11	72	70	0.002
Sacramento River Deep Water Channel, Light 55	261	23.2	23.9	7.1	8.2	7.64	8.13	84	66	0.002
DIEPAMHR + 25 ppb PBO	345	23.3	23.8	7.0	8.1	7.63	8.10	-	-	-
Low EC Control @ 112.0 µS/cm + 25 ppb PBO	130	23.3	23.8	7.3	8.0	7.23	7.71	-	-	-
High EC Control @ 18.56 mS/cm + 25 ppb PBO	17595	23.3	23.8	6.9	7.9	7.53	7.98	-	-	-
High EC Control @ 23.47 mS/cm + 25 ppb PBO	21850	23.3	23.8	6.3	7.9	7.44	7.94	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	4193	23.3	23.9	6.2	8.3	7.41	7.91	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	6575	23.3	23.8	6.9	8.4	7.64	7.97	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	16945	23.3	23.7	6.9	8.0	7.56	7.84	-	-	-
Carquinez Strait, West of Benecia army dock (405) + 25 ppb PBO	21255	23.3	23.8	6.8	8.0	7.55	7.86	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	351	23.3	23.7	7.2	8.3	7.57	8.06	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	138	23.3	23.7	6.8	8.5	7.45	7.96	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	387	23.2	23.7	6.8	8.3	7.49	8.09	-	-	-
Confluence of Lindsey Sl. and Cache Sl. + 25 ppb PBO	151	23.2	23.7	7.2	8.2	7.55	8.06	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	251	23.2	23.6	6.7	8.5	7.54	8.04	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	176	23.2	23.6	7.1	8.2	7.66	8.07	-	-	-
Sacramento River Deep Water Channel, Light 55 + 25 ppb PBO	258	23.3	23.5	7.0	8.4	7.60	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 B 92-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 7/23/09 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 7/22/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	100	0.0	100	0.0	NS
Low EC Control @ 123.2 µS/cm	100	0.0	100	0.0	NS
High EC Control @ 23.53 mS/cm	100	0.0	100	0.0	NS
Sacramento River at Hood DWR Station [3]	100	0.0	100	0.0	NS
Napa River, near Napa Park Blvd. [4]	98	2.5	98	2.5	NS
Suisun Slough at Rush Ranch	98	2.5	98	2.5	NS
Rough and Ready Island, DWR Station, Stockton	100	0.0	100	0.0	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.070	0.007	0.049	0.007	NS
Low EC Control @ 123.2 µS/cm	0.668	0.015	0.052	0.005	NS
High EC Control @ 23.53 mS/cm	0.042*	0.011	0.035	0.005	NS
Sacramento River at Hood DWR Station [3]	0.076	0.007	0.061	0.008	NS
Napa River, near Napa Park Blvd. [4]	0.054	0.010	0.045	0.005	NS
Suisun Slough at Rush Ranch	0.105	0.008	0.103	0.011	NS
Rough and Ready Island, DWR Station, Stockton	0.111	0.002	0.106	0.013	NS

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

*: $P < 0.05$

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

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

4. This high conductivity sample was compared to the High EC Control.

Table B 92-2. Summary of 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 7/22/09.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)[1]	Temp (^{o}C)	pH	DO (mg/L)			
Sacramento River at Hood DWR Station	-	23.3	7.31	8.0	17.3	0.13	-
Napa River, near Napa Park Blvd.	-	23.9	7.61	7.4	24.0	0.01	-
Suisun Slough at Rush Ranch	-	18.7	7.29	5.9	26.2	0.06	-
Rough and Ready Island, DWR Station, Stockton	-	26.6	7.44	5.6	7.8	0.02	-

1. Field EC meter broken and SC measurements were unable to be obtained.

Table B 92-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 7/23/09 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 7/22/09.

Treatment	Laboratory Chemistry							Hardness (mg/L as $CaCO_3$)	Alkalinity (mg/L as $CaCO_3$)	Unionized Ammonia $(mg/L)^2$
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	339	22.3	23.4	7.3	8.8	7.41	8.11	100	58	-
Low EC Control @ 123.2 µS/cm	132	22.3	23.6	7.2	8.5	6.90	7.90	36	24	-
High EC Control @ 23.53 mS/cm	21835	22.2	23.2	6.7	8.6	7.30	7.89	2720	78	-
Sacramento River at Hood DWR Statio	135	22.0	23.2	7.3	8.8	7.17	8.12	48	50	0.008
Napa River, near Napa Park Blvd.	21275	22.4	23.3	6.7	8.7	7.44	7.84	2760	134	0.000
Suisun Slough at Rush Ranch	8165	22.3	23.2	7.2	8.8	7.60	8.02	1160	138	0.001
Rough and Ready Island, DWR Station, Stockton	444	22.1	23.0	7.0	8.6	7.48	8.08	116	80	0.001
DIEPAMHR + 25 ppb PBO	333	22.1	23.2	7.4	8.6	7.40	8.14	-	-	-
Low EC Control @ 123.2 µS/cm + 25 ppb PBO	130	22.1	23.3	7.2	8.6	7.02	7.87	-	-	-
High EC Control @ 23.53 mS/cm + 25 ppb PBO	21655	22.3	23.2	6.5	8.5	7.31	7.90	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	134	22.2	23.3	6.7	8.8	7.20	8.02	-	-	-
Napa River, near Napa Park Blvd. + 25 ppb PBO	20890	22.4	23.9	6.2	8.6	7.47	7.91	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	4383	22.2	23.4	7.1	8.8	7.58	8.15	-	-	-
Rough and Ready Island, DWR Station, Stockton + 25 ppb PBO	4401	22.2	23.5	7.1	8.6	7.48	7.98	-	-	-

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

Table B 93-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 8/6/09 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 8/4/09-8/5/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	98	2.5	98	2.5	NS
Low EC Control @ 140 µS/cm	93	4.8	98	2.5	NS
High EC Control @ 16.00 mS/cm	97	2.8	100	0.0	NS
High EC Control @ 20.00 mS/cm	98	2.5	85	9.6	NS
Suisun Bay off Chipps Island (508)	100	0.0	100	0.0	NS
Rough and Ready DWR station, Stockton	92	5.3	90	4.1	NS
Sacramento River at Hood DWR Station[3]	93	4.8	98	2.5	NS
Suisun Slough at Rush Ranch	100	0.0	98	2.5	NS
Carquinez Strait, West of Benicia army dock (405)[5]	100	0.0	84	3.2	S* (84%)
Montezuma Slough at Nurse Slough (609)	100	0.0	97	3.1	NS
Grizzly Bay at Dolphin (602)[4]	88	4.5	100	0.0	NS
227 080409 (Bottle Blank)	97	2.8	-	-	NA
447 080409 (Trip Blank)	98	2.5	-	-	NA
337 080509 (Trip Blank)	98	2.5	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.053	0.008	0.048	0.007	NS
Low EC Control @ 140 µS/cm	0.055	0.004	0.032	0.005	S* (58%)
High EC Control @ 16.00 mS/cm	0.035	0.004	0.031*	0.003	NS
High EC Control @ 20.00 mS/cm	0.034*	0.003	0.010**	0.002	S** (29%)
Suisun Bay off Chipps Island (508)	0.064	0.015	0.055	0.006	NS
Rough and Ready DWR station, Stockton	0.089	0.009	0.074	0.009	NS
Sacramento River at Hood DWR Station[3]	0.053	0.002	0.056	0.003	NS
Suisun Slough at Rush Ranch	0.064	0.004	0.069	0.003	NS
Carquinez Strait, West of Benicia army dock (405)[5]	0.049	0.002	0.031	0.007	S* (63%)
Montezuma Slough at Nurse Slough (609)	0.077	0.006	0.066	0.011	NS
Grizzly Bay at Dolphin (602)[4]	0.054	0.006	0.037	0.005	NS
227 080409 (Bottle Blank)	0.059	0.008	-	-	NA
447 080409 (Trip Blank)	0.051	0.007	-	-	NA
337 080509 (Trip Blank)	0.041	0.006	-	-	NA

1. Data were analyzed using USEPA standard statistical protocols. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.
*: $P < 0.05$
**: $P < 0.01$
2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.
3. This low conductivity sample was compared to the Low EC Control.

4. This high conductivity sample was compared to the High EC Control @ 16.00 mS/cm.
5. These high conductivity samples were compared to the High EC Control @ 20.00 mS/cm.

Table B 93-2. Summary of 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 8/4/09 - 8/5/09.

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)	6410	19.8	7.78	9.4	28.7	0.06	0.001
Rough and Ready DWR station, Stockton	423	25.7	7.44	7.4	6.2	0.08	0.001
Sacramento River at Hood DWR Station	168	22.3	7.40	9.6	7.4	0.22	0.002
Suisun Slough at Rush Ranch	10490	17.3	7.28	7.3	22.6	0.09	0.000
Carquinez Strait, West of Benicia army dock (405)	18600	20.4	7.49	9.1	39.4	0.08	0.001
Montezuma Slough at Nurse Slough (609)	8990	20.9	7.44	8.6	19.2	0.04	0.000
Grizzly Bay at Dolphin (602)	15660	20.4	7.74	8.8	124.3	0.13	0.002
227 080409 (Bottle Blank)	-	-	-	-	-	-	-
447 080409 (Trip Blank)	-	-	-	-	0.3	0.03	-
337 080509 (Trip Blank)	-	-	-	-	0.3	0.04	-

Table B 93-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 8/6/09 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 8/4/09 - 8/5/09.

Treatment	Laboratory Chemistry								Hardness (mg/L as $CaCO_3$)	Alkalinity (mg/L as $CaCO_3$)	Unionized Ammonia (mg/L)[1]
	SC (uS/cm)	EC (uS/cm)	Min Temp (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	346.2	335	22.5	24.0	7.4	8.3	7.55	8.12	76	58	-
Low EC Control @ 140 µS/cm	154.05	150	22.5	24.5	7.3	8.5	7.30	7.92	40	22	-
High EC Control @ 16.00 mS/cm	15680	15265	22.7	24.4	7.1	7.9	7.60	7.97	1760	76	-
High EC Control @ 20.00 mS/cm	19785	19225	22.4	22.7	7.0	8.0	7.52	7.95	2320	82	-
Suisun Bay off Chipps Island (508)	6000	5845	22.8	23.1	6.9	8.3	7.40	7.85	660	56	0.002
Rough and Ready DWR station, Stockton	428.4	418	23.1	24.4	7.2	8.4	7.80	7.99	100	76	0.003
Sacramento River at Hood DWR Station	157.4	153	22.9	23.1	7.0	8.4	7.32	7.91	52	64	0.008
Suisun Slough at Rush Ranch	10065	9800	23.1	24.2	7.2	8.7	7.71	8.12	1240	146	0.002
Carquinez Strait, West of Benicia army dock (405)	18480	18015	23.1	24.3	6.9	7.7	7.50	7.81	2120	82	0.002
Montezuma Slough at Nurse Slough (609)	8780	4613	22.7	23.1	7.1	8.1	7.58	7.96	1040	90	0.001
Grizzly Bay at Dolphin (602)	15565	15180	22.7	23.1	7.0	8.1	7.50	7.85	1800	76	0.003
227 080409 (Bottle Blank)	357.5	1825	23.1	24.3	7.4	8.4	7.60	8.18	100	68	-
447 080409 (Trip Blank)	353.75	344	23.2	24.1	7.3	8.7	7.70	8.03	108	64	0.001
337 080509 (Trip Blank)	365.75	357	23.4	24.0	7.5	8.5	7.72	8.04	100	58	0.002
DIEPAMHR + 25 ppb PBO	352.9	341	23.2	23.2	7.4	8.2	7.54	8.06	-	-	-
Low EC Control @ 140 µS/cm + 25 ppb PBO	162.5	156	23.1	23.2	7.0	8.4	7.33	8.07	-	-	-
High EC Control @ 16.00 mS/cm + 25 ppb PBO	14729	14265	23.1	23.2	6.9	8.0	7.50	7.97	-	-	-
High EC Control @ 20.00 mS/cm + 25 ppb PBO	19680	18875	22.9	23.1	6.9	8.0	7.46	7.99	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	6185	5965	23.2	23.8	7.3	8.3	7.40	7.87	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	435.55	422	23.2	23.7	7.2	8.4	7.68	8.01	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	169.45	165	23.2	24.1	7.0	8.3	7.36	7.98	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	10100	9865	22.3	23.3	7.0	8.2	7.75	8.10	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	18285	17840	22.5	23.3	6.8	8.0	7.50	7.79	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	9185	8940	23.3	23.9	7.1	8.0	7.54	7.87	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	15385	14795	22.0	23.4	6.8	8.0	7.49	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 B 94-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 8/7/09 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 8/6/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	100	0.0	95	2.9	NS
Low EC Control @ 151.9 µS/cm	92	5.3	98	2.5	NS
Sacramento R. Deep Water Channel, Light 55	98	2.5	100	0.0	NS
Upper Cache Slough at mouth of Ulatis Creek[3]	92	8.3	98	2.5	NS
Confluence of Lindsey Sl. And Cache Sl.[3]	95	2.9	97	2.8	NS
Sacramento River at tip of Grand Island (711)[3]	79	4.1	80	12.2	NS
San Joaquin River at Potato Slough (815)	100	0.0	100	0.0	NS
Old River, western arm at railroad bridge (902)	98	2.5	100	0.0	NS
Old River at mouth of Holland Cut (915)	95	3.1	100	0.0	NS
Field Dup: Sacramento R. Deep Water Channel, Light 55	98	2.5	-	-	NA
557 080609 (Trip Blank)	95	2.9	-	-	NA
Field Dup.: Old River at mouth of Holland Cut (915)	98	2.5	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.033	0.011	0.045	0.009	NS
Low EC Control @ 151.9 µS/cm	0.033	0.005	0.031	0.005	NS
Sacramento R. Deep Water Channel, Light 55	0.079	0.005	0.076	0.006	NS
Upper Cache Slough at mouth of Ulatis Creek[3]	0.052	0.007	0.061	0.004	NS
Confluence of Lindsey Sl. And Cache Sl.[3]	0.060	0.006	0.050	0.008	NS
Sacramento River at tip of Grand Island (711)[3]	0.021	0.006	0.044	0.011	NS
San Joaquin River at Potato Slough (815)	0.072	0.006	0.076	0.008	NS
Old River, western arm at railroad bridge (902)	0.069	0.004	0.074	0.008	NS
Old River at mouth of Holland Cut (915)	0.075	0.006	0.064	0.003	NS
Field Dup: Sacramento R. Deep Water Channel, Light 55	0.091	0.006	-	-	NA
557 080609 (Trip Blank)	0.050	0.002	-	-	NA
Field Dup.: Old River at mouth of Holland Cut (915)	0.082	0.007	-	-	NA

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

*: $P < 0.05$

**: $P < 0.01$

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

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

Table B 94-2. Summary of 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 8/6/09.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (^{o}C)	pH	DO (mg/L)			
Sacramento R. Deep Water Channel, Light 55	258	20.7	8.15	9.3	32.0	0.06	0.003
Upper Cache Slough at mouth of Ulatis Creek	182	19.3	7.63	10.4	65.6	0.04	0.001
Confluence of Lindsey Sl. And Cache Sl.	167	19.8	7.55	9.5	41.1	0.05	0.001
Sacramento River at tip of Grand Island (711)	143	21.9	7.50	8.8	6.5	0.07	0.001
San Joaquin River at Potato Slough (815)	313	21.7	7.62	9.2	3.4	0.00	0.000
Old River, western arm at railroad bridge (902)	599	22.7	7.52	8.7	3.4	0.01	0.000
Old River at mouth of Holland Cut (915)	681	22.1	7.87	9.0	3.5	0.00	0.000
Field Dup: Sacramento R. Deep Water Channel, Light 55	258	20.7	8.15	9.3	32.3	0.04	0.002
557 080609 (Trip Blank)	-	-	-	-	0.3	0.00	-
Field Dup.: Old River at mouth of Holland Cut (915)	681	22.1	7.87	9.0	3.2	0.00	0.000

Table B 94-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 8/7/09 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 8/6/09.

Treatment	Laboratory Chemistry								Hardness (mg/L as $CaCO_3$)	Alkalinity (mg/L as $CaCO_3$)	Unionized Ammonia (mg/L)[1]
	SC (uS/cm)	EC (uS/cm)	Min Temp (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	356	347	23.6	23.7	7.2	8.1	7.56	8.14	76	60	-
Low EC Control @ 151.9 µS/cm	153	149	23.6	23.7	7.3	8.2	7.27	7.91	40	24	-
Sacramento R. Deep Water Channel, Light 55	270	263	23.6	23.6	6.8	8.4	7.55	8.10	92	70	0.003
Upper Cache Slough at mouth of Ulatis Creek	176	170	23.6	23.7	7.2	8.3	7.58	8.11	60	62	0.002
Confluence of Lindsey Sl. And Cache Sl.	164	159	23.1	23.5	7.1	8.2	7.50	8.06	60	60	0.003
Sacramento River at tip of Grand Island (711)	143	138	23.2	23.5	7.0	8.3	7.46	8.02	44	52	0.003
San Joaquin River at Potato Slough (815)	299	291	23.6	23.7	7.1	8.3	7.47	8.01	60	52	0.000
Old River, western arm at railroad bridge (902)	671	652	23.5	23.6	7.1	8.4	7.47	8.07	104	50	0.001
Old River at mouth of Holland Cut (915)	539	526	23.4	23.7	7.2	8.5	7.47	8.01	88	54	0.000
Field Dup: Sacramento R. Deep Water Channel, Light 55	271	263	23.5	23.5	7.0	8.4	7.57	8.11	76	68	0.002
557 080609 (Trip Blank)	349	339	23.5	23.8	7.1	8.2	7.56	8.09	104	56	0.000
Field Dup.: Old River at mouth of Holland Cut (915)	678	658	23.5	23.5	7.1	8.3	7.49	8.06	96	52	0.000
DIEPAMHR + 25 ppb PBO	351	339	22.1	23.8	7.2	8.2	7.55	8.14	-	-	-
Low EC Control @ 151.9 µS/cm + 25 ppb PBO	152	146	22.3	23.5	7.2	8.3	7.25	7.87	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	267	257	22.6	23.5	7.2	8.8	7.61	8.11	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	177	171	22.5	23.6	7.2	9.2	7.54	8.12	-	-	-
Confluence of Lindsey Sl. And Cache Sl. + 25 ppb PBO	164	159	22.8	23.7	7.2	8.4	7.53	8.10	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	142	139	22.9	23.8	6.9	8.2	7.51	8.12	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	345	337	23.4	23.8	7.1	8.3	7.47	8.08	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	670	654	23.3	23.8	7.1	8.5	7.49	8.07	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	527	512	23.2	23.9	7.0	8.3	7.46	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 B 95-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 8/20/09 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 8/18/09 - 8/19/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	100	0.0	98	2.5	NS
Low EC @ 174.9 uS/cm	100	0.0	94	3.3	NS
High EC @ 11.88 mS/cm	100	0.0	98	2.5	NS
High EC @ 18.4 mS/cm	95	2.9	95	5.0	NS
Suisun Slough at Rush Ranch[4]	100	0.0	95	4.5	NS
Rough and Ready DWR station, Stockton	100	0.0	98	2.5	NS
Sacramento River at Hood DWR Station[3]	95	3.1	97	3.1	NS
Grizzly Bay at Dolphin (602)[5]	94	5.6	100	0.0	NS
Suisun Bay off Chipps Island (508)	98	2.5	98	2.5	NS
Montezuma Slough at Nurse Slough (609)[4]	100	0.0	100	0.0	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.045	0.005	0.027	0.008	NS
Low EC @ 174.9 uS/cm	0.024	0.001	0.027	0.007	NS
High EC @ 11.88 mS/cm	0.036	0.006	0.027	0.006	NS
High EC @ 18.4 mS/cm	0.019	0.004	0.030	0.004	NS
Suisun Slough at Rush Ranch[4]	0.089	0.005	0.076	0.007	NS
Rough and Ready DWR station, Stockton	0.100	0.012	0.066	0.007	S* (66%)
Sacramento River at Hood DWR Station[3]	0.088	0.006	0.079	0.006	NS
Grizzly Bay at Dolphin (602)[5]	0.060	0.005	0.061	0.007	NS
Suisun Bay off Chipps Island (508)	0.058	0.006	0.070	0.011	NS
Montezuma Slough at Nurse Slough (609)[4]	0.075	0.005	0.087	0.006	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.
*: $P < 0.05$
2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.
3. This low conductivity sample was compared to the Low EC Control.
4. These high conductivity samples were compared to the High EC Control @ 11.88 mS/cm.
5. This high conductivity sample was compared to the High EC Control @ 18.4 mS/cm.

Table B 95-2. Summary of 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 8/18/09 - 8/19/09.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Suisun Slough at Rush Ranch	11440	22.1	7.37	6.5	9.4	0.06	0.000
Rough and Ready DWR station, Stockton	490	26.7	7.70	6.7	6.9	0.13	0.004
Sacramento River at Hood DWR Station	167	23.9	7.53	7.9	10.4	0.02	0.000
Grizzly Bay at Dolphin (602)	17960	20.5	7.68	9.0	35.4	0.03	0.000
Suisun Bay off Chipps Island (508)	7580	20.4	7.50	9.1	10.6	0.00	0.000
Montezuma Slough at Nurse Slough (609)	10440	21.6	7.46	8.2	31.4	0.04	0.000

Table B 95-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 8/20/09 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 8/18/09 - 8/19/09.

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 (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	345	23.3	24.0	7.4	8.2	7.71	8.13	104	60	-
Low EC @ 174.9 uS	182	23.3	23.8	7.2	8.4	7.38	7.96	52	30	-
High EC @ 11.88 mS	11180	23.3	23.8	7.2	8.2	7.53	7.97	1440	72	-
High EC @ 18.4 mS	17330	23.3	23.6	6.6	8.3	7.55	8.01	2080	78	-
Suisun Slough at Rush Ranch	10725	23.3	23.6	7.1	8.4	7.84	8.02	1520	152	0.002
Rough and Ready DWR station, Stockton	475	23.3	23.6	7.5	8.8	7.85	8.04	116	84	0.006
Sacramento River at Hood DWR Station	171	23.2	23.4	7.3	8.7	7.65	7.99	60	68	0.001
Grizzly Bay at Dolphin (602)	16675	23.0	23.6	7.0	8.0	7.45	7.80	2040	80	0.001
Suisun Bay off Chipps Island (508)	7085	23.1	23.4	7.3	8.4	7.52	7.85	840	66	0.000
Montezuma Slough at Nurse Slough (609)	9975	23.1	24.3	7.3	8.6	7.61	7.89	1200	94	0.001
DIEPAMHR + 25 ppb PBO	350	23.1	23.6	7.3	8.3	7.72	8.13	-	-	-
Low EC @ 174.9 uS/cm + 25 ppb PBO	184	23.1	23.6	7.2	8.6	7.41	7.92	-	-	-
High EC @ 11.88 mS/cm + 25 ppb PBO	11180	23.1	23.5	6.9	8.4	7.59	7.98	-	-	-
High EC @ 18.4 mS/cm + 25 ppb PBO	17315	23.1	23.5	6.7	8.4	7.55	8.01	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	10555	23.2	23.4	7.2	8.4	7.68	8.03	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	481	23.5	23.9	7.5	8.7	7.74	8.05	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	175	23.4	23.5	7.2	8.6	7.66	7.98	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	16580	23.4	23.6	7.1	8.5	7.51	7.81	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	7050	23.4	23.9	7.3	8.4	7.56	7.90	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	9990	23.4	24.0	7.3	8.4	7.67	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 B 96-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 8/21/09 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 8/20/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	100	0.0	95	2.9	NS
Low EC Control @ 164.4 µS/cm	100	0.0	94	6.3	NS
Sacramento River Deep Water Channel, Light 55	98	2.5	100	0.0	NS
Upper Cache Slough at mouth of Ulatis Creek[3]	90	7.1	98	2.5	NS
Confluence of Lindsey Sl. and Cache Sl.[3]	98	2.5	100	0.0	NS
Sacramento River at tip of Grand Island (711)[3]	98	2.5	98	2.5	NS
San Joaquin River at Potato Slough (815)	100	0.0	93	7.5	NS
Old River, western arm at railroad bridge (902)	95	2.9	100	0.0	NS
Old River at mouth of Holland Cut (915)	84	15.6	100	0.0	NS
Field Dup: Confluence of Lindsey Sl. and Cache Sl.[3]	98	2.5	-	-	NA
Field Dup: Old River at mouth of Holland Cut (915)	98	2.5	-	-	NA
Bottle Blank: DIEPAMHR	98	2.5	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.030	0.007	0.038	0.002	NS
Low EC Control @ 164.4 µS/cm	0.026	0.005	0.032	0.003	NS
Sacramento River Deep Water Channel, Light 55	0.114	0.007	0.114	0.015	NS
Upper Cache Slough at mouth of Ulatis Creek[3]	0.082	0.003	0.087	0.004	NS
Confluence of Lindsey Sl. and Cache Sl.[3]	0.101	0.008	0.072	0.008	S* (71%)
Sacramento River at tip of Grand Island (711)[3]	0.079	0.007	0.073	0.013	NS
San Joaquin River at Potato Slough (815)	0.112	0.003	0.065	0.018	S* (58%)
Old River, western arm at railroad bridge (902)	0.083	0.017	0.080	0.012	NS
Old River at mouth of Holland Cut (915)	0.084	0.016	0.089	0.004	NS
Field Dup: Confluence of Lindsey Sl. and Cache Sl.[3]	0.092	0.007	-	-	NA
Field Dup: Old River at mouth of Holland Cut (915)	0.113	0.016	-	-	NA
Bottle Blank: DIEPAMHR	0.033	0.003	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard statistical protocols.

*: $P < 0.05$

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

3. These low conductivity samples were compared to the Low EC Control.

Table B 96-2. Summary of 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 8/20/09.

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 Deep Water Channel, Light 55	258	21.4	7.97	8.7	28.7	0.06	0.002
Upper Cache Slough at mouth of Ulatis Creek	220	20.1	7.72	9.0	66.9	0.07	0.001
Confluence of Lindsey Sl. and Cache Sl.	209	20.6	7.81	9.0	35.4	0.08	0.002
Sacramento River at tip of Grand Island (711)	255	22.5	7.52	8.5	8.6	0.10	0.001
San Joaquin River at Potato Slough (815)	394	22.7	7.50	8.6	4.1	0.03	0.000
Old River, western arm at railroad bridge (902)	720	23.2	7.92	8.5	3.5	0.00	0.000
Old River at mouth of Holland Cut (915)	643	24.2	7.74	8.3	3.7	0.00	0.000
Field Dup: Confluence of Lindsey Sl. and Cache Sl.	209	20.6	7.81	9.0	33.9	0.11	0.003
Field Dup: Old River at mouth of Holland Cut (915)	643	24.2	7.74	8.3	3.7	0.00	0.000

Table B 96-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 8/21/09 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 8/20/09.

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	24.5	24.5	7.2	8.1	7.53	8.09	104	60	-
Low EC Control @ 164.4 µS/cm	180	24.2	24.4	7.1	8.7	7.28	7.81	52	28	-
Sacramento River Deep Water Channel, Light 55	271	24.0	24.6	7.1	8.7	7.55	8.05	80	74	0.003
Upper Cache Slough at mouth of Ulatis Creek	190	24.0	24.4	7.1	8.8	7.57	8.04	68	68	0.004
Confluence of Lindsey Sl. and Cache Sl.	185	23.9	24.0	7.1	8.4	7.51	8.04	68	68	0.004
Sacramento River at tip of Grand Island (711)	174	23.9	24.4	7.0	8.6	7.43	7.97	60	68	0.005
San Joaquin River at Potato Slough (815)	362	24.1	24.4	7.1	8.7	7.50	7.97	76	60	0.001
Old River, western arm at railroad bridge (902)	668	24.0	24.4	7.2	8.4	7.73	8.07	100	56	0.000
Old River at mouth of Holland Cut (915)	589	23.9	24.4	6.5	8.5	7.49	8.01	100	58	0.000
Field Dup: Confluence of Lindsey Sl. and Cache Sl.	187	23.9	24.2	7.2	8.7	7.64	8.16	68	66	0.007
Field Dup: Old River at mouth of Holland Cut (915)	587	23.9	24.5	7.1	8.7	7.46	7.97	96	56	0.000
Bottle Blank: DIEPAMHR	355	23.9	24.3	7.3	8.3	7.56	8.08	116	58	0.001
DIEPAMHR + 25 ppb PBO	351	23.9	24.0	7.1	8.3	7.59	8.09	-	-	-
Low EC Control @ 164.4 µS/cm + 25 ppb PBO	179	23.9	24.0	7.1	8.5	7.26	7.82	-	-	-
Sacramento River Deep Water Channel, Light 55 + 25 ppb PBO	267	23.9	24.2	7.0	8.7	7.62	8.05	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	189	23.9	24.2	7.3	8.5	7.61	8.05	-	-	-
Confluence of Lindsey Sl. and Cache Sl. + 25 ppb PBO	186	24.0	24.2	7.3	8.4	7.52	8.05	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	170	24.1	24.1	6.8	8.7	7.44	8.14	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	364	24.2	24.3	7.1	8.4	7.56	7.97	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	667	24.3	24.3	7.0	8.4	7.52	8.07	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	587	24.1	24.4	7.5	8.3	7.51	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 B 97-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 9/03/09 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 9/01/09 - 9/02/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	98	2.5	100	0.0	NS
Low EC Control @ 180.1 uS/cm	100	0.0	100	0.0	NS
Sacramento R. Deep Water Channel, Light 55	100	0.0	98	2.5	NS
Upper Cache Slough at mouth of Ulatis Creek[3]	100	0.0	100	0.0	NS
Confluence of Lindsey Sl. And Cache Sl.[3]	93	4.4	98	2.5	NS
Sacramento River at tip of Grand Island (711)[3]	98	2.5	90	4.1	NS
San Joaquin River at Potato Slough (815)	100	0.0	100	0.0	NS
Old River, western arm at railroad bridge (902)	100	0.0	100	0.0	NS
Old River at mouth of Holland Cut (915)	100	0.0	98	2.3	NS
Suisun Bay off Chipps Island (508)	100	0.0	100	0.0	NS
337 (Trip Blank)	98	2.5	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.077	0.005	0.062	0.002	S* (81%)
Low EC Control @ 180.1 uS/cm	0.061	0.006	0.064	0.003	NS
Sacramento R. Deep Water Channel, Light 55	0.117	0.008	0.117	0.009	NS
Upper Cache Slough at mouth of Ulatis Creek[3]	0.121	0.004	0.096	0.003	S** (79%)
Confluence of Lindsey Sl. And Cache Sl.[3]	0.138	0.002	0.100	0.005	S*** (72%)
Sacramento River at tip of Grand Island (711)[3]	0.111	0.010	0.108	0.003	NS
San Joaquin River at Potato Slough (815)	0.120	0.004	0.105	0.005	S* (88%)
Old River, western arm at railroad bridge (902)	0.112	0.027	0.106	0.007	NS
Old River at mouth of Holland Cut (915)	0.136	0.011	0.102	0.008	S* (75%)
Suisun Bay off Chipps Island (508)	0.105	0.016	0.091	0.007	NS
337 (Trip Blank)	0.062	0.004	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed usign USEPA standard statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

***: $P < 0.001$

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

3. These low conductivity samples were compared to the Low EC Control.

Table B 97-2. Summary of 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 9/01/09 - 9/02/09.

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	357	21.8	8.03	8.4	32.6	0.04	0.002
Upper Cache Slough at mouth of Ulatis Creek	227	20.6	7.78	8.7	45.9	0.07	0.002
Confluence of Lindsey Sl. And Cache Sl.	417	21.3	7.64	8.6	23.3	0.09	0.002
Sacramento River at tip of Grand Island (711)	197	22.1	7.63	8.3	4.0	0.09	0.002
San Joaquin River at Potato Slough (815)	333	23.3	7.36	8.2	2.3	0.02	0.000
Old River, western arm at railroad bridge (902)	853	24.5	8.21	8.3	1.8	0.00	0.000
Old River at mouth of Holland Cut (915)	846	25.1	7.77	8.1	1.9	0.00	0.000
Suisun Bay off Chipps Island (508)	6180	21.2	7.69	8.5	10.9	0.07	0.001
337 (Trip Blank)	-	-	-	-	0.3	0.05	-

Table B 97-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 9/03/09 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 9/01/09 - 9/02/09.

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 (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	347	22.7	24.3	7.1	8.2	7.60	8.14	104	56	-
Low EC Control @ 180.1 uS/cm	939	22.8	24.3	7.4	8.4	7.55	8.02	-	-	-
Sacramento R. Deep Water Channel, Light 55	322	22.9	24.4	7.0	8.9	7.86	8.15	88	80	0.003
Upper Cache Slough at mouth of Ulatis Creek	208	22.9	24.4	7.4	8.6	7.99	8.21	72	78	0.005
Confluence of Lindsey Sl. And Cache Sl.	186	22.9	24.2	7.0	8.8	7.80	8.19	54	72	0.007
Sacramento River at tip of Grand Island (711)	177	22.9	24.3	7.0	8.7	7.81	8.02	76	70	0.005
San Joaquin River at Potato Slough (815)	318	22.9	24.2	7.3	8.4	7.81	8.06	72	68	0.001
Old River, western arm at railroad bridge (902)	678	22.9	24.3	7.0	8.6	7.67	8.21	104	62	0.000
Old River at mouth of Holland Cut (915)	575	22.9	24.3	7.0	8.5	7.66	8.10	96	62	0.000
Suisun Bay off Chipps Island (508)	5725	22.9	24.3	6.7	8.7	7.46	7.98	640	68	0.003
337 (Trip Blank)	336	22.9	24.3	7.5	8.7	7.74	8.15	108	54	0.003
DIEPAMHR + 25 ppb PBO	337	22.9	24.2	7.3	8.3	7.63	8.14	-	-	-
Low EC Control @ 180.1 uS/cm + 25 ppb PBO	961	22.9	24.3	7.6	8.5	7.60	7.95	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	321	22.9	24.4	7.2	8.5	7.96	8.14	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	208	22.9	24.4	7.3	8.8	7.96	8.17	-	-	-
Confluence of Lindsey Sl. And Cache Sl. + 25 ppb PBO	188	23.0	24.4	7.1	8.7	7.85	8.16	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	177	23.0	24.5	6.8	8.8	7.78	8.15	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	316	23.1	24.3	7.5	8.9	7.74	8.08	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	679	23.3	24.3	7.0	8.6	7.61	8.21	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	577.5	23.3	24.4	7.0	8.5	7.65	8.08	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	5725	23.1	24.3	6.9	8.7	7.48	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 B 98-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 9/04/09 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 9/02/09 - 9/03/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	100	0.0	88	2.5	S* (88%)
Low EC Control @ 177.7 uS/cm	98	2.5	98	2.5	NS
High EC Control @ 13.40 mS/cm	100	0.0	88	12.5	NS
High EC Control @ 17.40 mS/cm	100	0.0	92	5.3	NS
High EC Control @ 24.10 mS/cm	81	7.9	48	19.7	NS
Montezuma Slough at Nurse Slough (609)	100	0.0	100	0.0	NS
Grizzly Bay at Dolphin (602) [5]	100	0.0	88	6.3	NS
Carquinez Strait, West of Benicia army dock (405) [6]	85	11.9	48	9.3	S* (56%)
Sacramento River at Hood DWR Station [3]	100	0.0	95	5.0	NS
Rough and Ready Island, DWR Station, Stockton	100	0.0	98	2.5	NS
Suisun Slough at Rush Ranch [4]	100	0.0	88	12.5	NS
Field Dup: Sacramento River at Hood DWR Station [3]	95	2.9	-	-	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.054	0.011	0.042	0.006	NS
Low EC Control @ 177.7 uS/cm	0.042	0.003	0.029	0.012	NS
High EC Control @ 13.40 mS/cm	0.036	0.003	0.033	0.003	NS
High EC Control @ 17.40 mS/cm	0.028*	0.002	0.020*	0.005	NS
High EC Control @ 24.10 mS/cm	0.025*	0.003	0.021*	0.003	NS
Montezuma Slough at Nurse Slough (609)	0.072	0.009	0.061	0.005	NS
Grizzly Bay at Dolphin (602) [5]	0.056	0.008	0.048	0.004	NS
Carquinez Strait, West of Benicia army dock (405) [6]	0.052	0.006	0.055	0.007	NS
Sacramento River at Hood DWR Station [3]	0.079	0.006	0.087	0.012	NS
Rough and Ready Island, DWR Station, Stockton	0.112	0.008	0.098	0.008	NS
Suisun Slough at Rush Ranch [4]	0.082	0.005	0.082	0.011	NS
Field Dup: Sacramento River at Hood DWR Station [3]	0.087	0.007	-	-	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard statistical protocols.
*: $P < 0.05$
2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.
3. This low conductivity sample was compared to the Low EC Control.
4. This high conductivity sample was compared to the High EC Control @ 13.40 mS/cm.
5. This high conductivity sample was compared to the High EC Control @ 17.40 mS/cm.
6. This high conductivity sample was compared to the High EC Control @ 24.10 mS/cm.

Table B 98-2. Summary of 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 9/02/09 - 9/03/09.

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)	11750	22.8	7.55	7.4	13.0	0.06	0.001
Grizzly Bay at Dolphin (602)	17400	21.7	7.70	8.7	12.7	0.04	0.001
Carquinez Strait, West of Benicia army dock (405)	23720	20.6	7.89	8.1	19.6	0.09	0.002
Sacramento River at Hood DWR Station	188	23.3	7.64	7.6	10.9	0.06	0.003
Rough and Ready Island, DWR Station, Stockton	576	26.9	7.61	7.4	8.4	0.00	0.000
Suisun Slough at Rush Ranch	12810	19.7	7.28	7.3	19.2	0.00	0.000
Field Dup: Sacramento River at Hood DWR Station	188	23.3	7.64	7.6	103.0	0.00	0.000

Table B 98-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 9/04/09 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 9/02/09 - 9/03/09.

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	23.1	24.2	7.5	8.3	7.65	8.10	104	56	-
Low EC Control @ 177.7 uS/cm	182	23.0	24.4	7.6	8.8	7.54	7.89	64	28	-
High EC Control @ 13.40 mS/cm	12695	22.9	24.3	7.4	8.0	7.56	7.98	1640	66	-
High EC Control @ 17.40 mS/cm	16660	22.8	24.3	7.2	8.1	7.64	7.98	1920	78	-
High EC Control @ 24.10 mS/cm	22370	22.7	23.9	7.1	7.6	7.67	7.99	2720	86	-
Montezuma Slough at Nurse Slough (609)	11120	22.7	23.8	7.3	8.3	7.71	7.88	1232	94	0.001
Grizzly Bay at Dolphin (602)	16210	22.7	24.2	7.2	8.1	7.59	7.79	1880	80	0.001
Carquinez Strait, West of Benicia army dock (405)	22980	22.7	23.8	6.6	8.1	7.33	7.78	2720	86	0.002
Sacramento River at Hood DWR Station	220	22.9	24.0	7.1	8.5	7.77	8.02	72	72	0.003
Rough and Ready Island, DWR Station, Stockton	575	22.9	24.0	7.3	8.5	7.86	7.99	132	82	0.000
Suisun Slough at Rush Ranch	12290	22.9	24.0	6.9	8.4	7.61	8.02	1520	152	0.000
Field Dup: Sacramento River at Hood DWR Station	210	22.8	24.7	7.1	8.7	7.68	7.91	72	70	0.000
DIEPAMHR + 25 ppb PBO	345	23.0	24.2	7.6	8.2	7.68	8.12	-	-	-
Low EC Control @ 177.7 uS/cm + 25 ppb PBO	187	23.0	23.8	7.5	8.5	7.57	7.88	-	-	-
High EC Control @ 13.40 mS/cm + 25 ppb PBO	12620	22.9	24.0	7.4	8.1	7.60	7.98	-	-	-
High EC Control @ 17.40 mS/cm + 25 ppb PBO	16560	22.9	24.3	7.2	8.0	7.60	7.98	-	-	-
High EC Control @ 24.10 mS/cm + 25 ppb PBO	23230	23.0	24.4	7.1	7.8	7.69	7.99	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	11250	22.9	24.1	7.2	8.2	7.70	7.88	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	16700	22.9	24.3	7.3	7.9	7.61	7.85	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	22355	22.8	24.1	6.6	7.7	7.53	7.81	-	-	-

Sacramento River at Hood DWR Station + 25 ppb PBO	224	22.8	24.1	7.0	8.5	7.78	8.06	-	-	-
Rough and Ready Island, DWR Station, Stockton + 25 ppb PBO	582	22.9	24.2	7.3	8.5	7.83	8.01	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	12245	22.9	24.3	7.2	8.3	7.55	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 B 99-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 9/17/09 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 9/15/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	100	0.0	100	0.0	NS
Low EC Control @ 193.1 µS/cm	100	0.0	95	5.0	NS
Upper Cache Slough at mouth of Ulatis Creek[3]	100	0.0	93	4.8	NS
Old River at mouth of Holland Cut (915)	98	2.5	100	0.0	NS
Sacramento River at tip of Grand Island (711)[3]	100	0.0	98	2.5	NS
Confluence of Lindsey Sl. And Cache Sl.[3]	100	0.0	100	0.0	NS
San Joaquin River at Potato Slough (815)	100	0.0	100	0.0	NS
Old River, western arm at railroad bridge (902)	100	0.0	98	2.3	NS
Sacramento R. Deep Water Channel, Light 55	100	0.0	98	2.5	NS
Field Dup.: Old River at mouth of Holland Cut (915)	100	0.0	-	-	NA
337 091509 Trip Blank	95	5.0	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.043	0.008	0.032	0.005	NS
Low EC Control @ 193.1 µS/cm	0.042	0.007	0.033	0.003	NS
Upper Cache Slough at mouth of Ulatis Creek[3]	0.089	0.006	0.101	0.011	NS
Old River at mouth of Holland Cut (915)	0.106	0.008	0.093	0.008	NS
Sacramento River at tip of Grand Island (711)[3]	0.100	0.005	0.075	0.009	S* (75%)
Confluence of Lindsey Sl. And Cache Sl.[3]	0.090	0.008	0.089	0.015	NS
San Joaquin River at Potato Slough (815)	0.101	0.006	0.069	0.012	NS
Old River, western arm at railroad bridge (902)	0.097	0.006	0.109	0.004	NS
Sacramento R. Deep Water Channel, Light 55	0.106	0.002	0.092	0.007	NS
Field Dup.: Old River at mouth of Holland Cut (915)	0.103	0.009	-	-	NA
337 091509 Trip Blank	0.051	0.010	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard statistical protocols.

*: $P < 0.05$

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

3. These low conductivity samples were compared to the Low EC Control.

Table B 99-2. Summary of 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 9/15/09.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (^{o}C)	pH	DO (mg/L)			
Upper Cache Slough at mouth of Ulatis Creek	245	21.1	7.77	8.5	33.7	0.04	0.001
Old River at mouth of Holland Cut (915)	677	23.4	7.63	8.2	2.4	0.01	0.000
Sacramento River at tip of Grand Island (711)	196	22.3	7.51	8.1	4.3	0.22	0.003
Confluence of Lindsey Sl. And Cache Sl.	234	20.8	7.79	8.4	25.0	0.09	0.002
San Joaquin River at Potato Slough (815)	449	22.6	7.53	8.2	3.4	0.04	0.001
Old River, western arm at railroad bridge (902)	780	23.0	8.00	8.9	2.2	0.00	0.000
Sacramento R. Deep Water Channel, Light 55	366	21.7	7.78	8.5	22.8	0.03	0.001
Field Dup.: Old River at mouth of Holland Cut (915)	677	23.4	7.63	8.2	2.6	0.00	0.000
337 091509 Trip Blank	-	-	-	-	0.3	0.03	-

Table B 99-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 9/17/09 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 9/15/09.

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	341	23.0	24.6	7.1	8.3	7.48	8.01	104	62	-
Low EC Control @ 193.1 µS/cm	192	22.9	24.7	7.1	8.5	7.28	7.86	56	34	-
Upper Cache Slough at mouth of Ulatis Creek	229	22.9	24.3	7.2	8.7	7.59	8.03	80	86	0.002
Old River at mouth of Holland Cut (915)	640	22.9	24.4	7.1	8.7	7.42	7.99	108	70	0.000
Sacramento River at tip of Grand Island (711)	194	22.9	24.2	7.0	8.8	7.51	7.90	68	74	0.008
Confluence of Lindsey Sl. And Cache Sl.	218	22.9	24.4	7.3	8.7	7.45	8.07	76	80	0.005
San Joaquin River at Potato Slough (815)	437	23.0	24.5	7.2	8.6	7.50	8.04	92	72	0.002
Old River, western arm at railroad bridge (902)	737	23.0	24.6	7.2	8.8	7.52	8.10	116	68	0.000
Sacramento R. Deep Water Channel, Light 55	343	22.9	24.5	7.2	8.8	7.62	8.15	96	86	0.002
Field Dup.: Old River at mouth of Holland Cut (915)	639	22.9	24.5	7.1	8.4	7.51	8.01	104	70	0.000
337 091509 Trip Blank	336	23.0	24.5	7.3	8.5	7.53	8.02	104	56	0.002
DIEPAMHR + 25 ppb PBO	340	23.1	24.1	7.2	8.3	7.42	8.04	-	-	-
Low EC Control @ 193.1 µS/cm + 25 ppb PBO	188	23.0	24.2	7.1	8.8	7.25	7.86	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	222	23.0	24.2	7.1	8.6	7.59	8.13	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	635	22.9	24.2	7.1	8.4	7.44	7.98	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	180	23.0	24.1	7.1	8.6	7.44	8.01	-	-	-
Confluence of Lindsey Sl. And Cache Sl. + 25 ppb PBO	204	23.2	24.3	7.2	8.5	7.54	8.02	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	439	23.0	24.3	7.2	8.4	7.49	8.00	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	732	23.2	24.2	7.2	8.8	7.60	8.00	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	344	23.0	24.4	7.2	8.2	7.63	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 B 100-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 9/18/09 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 9/16/09 - 9/17/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	91	3.0	97	2.8	NS
Low EC Control @ 193.9 µS/cm	94	3.4	100	0.0	NS
High EC Control @ 14.35 mS/cm	100	0.0	97	2.8	NS
High EC Control @ 18.20 mS/cm	98	2.5	100	0.0	NS
High EC Control @ 25.07 mS/cm	64	3.1	86	10.5	NS
Grizzly Bay at Dolphin (602)[5]	95	2.9	87*	2.4	NS
Suisun Bay off Chipps Island (508)	100	0.0	100	0.0	NS
Montezuma Slough at Nurse Slough (609)[4]	98	2.5	100	0.0	NS
Carquinez Strait, West of Benicia army dock (405)[6]	78	7.5	69	8.5	NS
Suisun Slough at Rush Ranch	98	2.5	100	0.0	NS
Sacramento River at Hood DWR Station[3]	98	2.5	100	0.0	NS
Rough and Ready DWR station, Stockton	98	2.5	100	0.0	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.060	0.011	0.070	0.006	NS
Low EC Control @ 193.9 µS/cm	0.059	0.004	0.076	0.010	NS
High EC Control @ 14.35 mS/cm	0.078	0.004	0.049*	0.004	S** (63%)
High EC Control @ 18.20 mS/cm	0.055	0.003	0.051*	0.002	NS
High EC Control @ 25.07 mS/cm	0.032*	0.004	0.051	0.012	NS
Grizzly Bay at Dolphin (602)[5]	0.067	0.001	0.049	0.007	NS
Suisun Bay off Chipps Island (508)	0.103	0.007	0.084	0.012	NS
Montezuma Slough at Nurse Slough (609)[4]	0.079	0.006	0.074	0.006	NS
Carquinez Strait, West of Benicia army dock (405)[6]	0.057	0.005	0.040	0.013	NS
Suisun Slough at Rush Ranch	0.099	0.004	0.082	0.003	S* (83%)
Sacramento River at Hood DWR Station[3]	0.104	0.002	0.081	0.003	S** (78%)
Rough and Ready DWR station, Stockton	0.135	0.004	0.117	0.003	S* (87%)

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard statistical protocols.
 *: $P < 0.05$
 **: $P < 0.01$
2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.
3. These treatments were compared to the Low EC Control.
4. These treatments were compared to the High EC Control @ 14.35 mS/cm.
5. These treatments were compared to the High EC Control @ 18.2 mS/cm.
6. These treatments were compared to the High EC Control @ 25.07 mS/cm.

Table B 100-2. Summary of 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 9/16/09 - 9/17/09.

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)	17710	20.6	7.46	8.7	21.1	0.08	0.001
Suisun Bay off Chipps Island (508)	6220	20.5	7.53	8.9	9.3	0.09	0.001
Montezuma Slough at Nurse Slough (609)	13010	21.8	7.44	8.0	23.5	0.15	0.001
Carquinez Strait, West of Benicia army dock (405)	24290	20.5	7.62	8.4	29.5	0.18	0.002
Suisun Slough at Rush Ranch	13730	18.7	7.13	7.4	40.8	0.19	0.001
Sacramento River at Hood DWR Station	192	22.0	7.49	8.0	9.6	0.32	0.004
Rough and Ready DWR station, Stockton	572	24.3	7.44	7.2	8.9	0.10	0.001

Table B 100-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 9/18/09 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 9/16/09 - 9/17/09.

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	22.7	23.9	7.6	8.7	7.66	8.02	104	62	-
Low EC Control @ 193.9 µS/cm	191	22.6	24.0	7.5	8.8	7.47	7.80	64	30	-
High EC Control @ 14.35 mS/cm	13350	22.4	24.0	7.2	8.8	7.63	7.76	1680	74	-
High EC Control @ 18.20 mS/cm	17265	22.5	24.4	7.2	8.4	7.61	7.77	2120	78	-
High EC Control @ 25.07 mS/cm	24035	22.6	24.3	6.8	8.5	7.63	7.77	2920	84	-
Grizzly Bay at Dolphin (602)	16905	22.5	24.4	7.2	8.8	7.63	7.79	2720	82	0.002
Suisun Bay off Chipps Island (508)	5890	22.4	24.3	7.3	8.7	7.60	7.90	720	74	0.003
Montezuma Slough at Nurse Slough (609)	12440	22.3	23.9	7.3	8.6	7.68	7.80	1480	116	0.003
Carquinez Strait, West of Benicia army dock (405)	23320	22.5	24.3	6.8	8.8	7.60	7.79	2080	92	0.004
Suisun Slough at Rush Ranch	13130	22.5	24.4	6.9	8.9	7.87	8.04	160	150	0.005
Sacramento River at Hood DWR Station	197	22.1	24.3	6.8	8.6	7.74	8.20	72	74	0.024
Rough and Ready DWR station, Stockton	555	22.1	24.2	7.4	8.9	7.72	8.09	120	82	0.006
DIEPAMHR	347	22.6	23.9	7.5	8.7	7.54	8.08	-	-	-
Low EC Control @ 193.9 µS/cm	201	22.6	23.8	7.4	8.8	7.42	7.87	-	-	-
High EC Control @ 14.35 mS/cm	13500	22.5	23.8	7.1	8.4	7.64	8.66	-	-	-
High EC Control @ 18.20 mS/cm	17295	22.5	23.7	7.1	8.4	7.67	7.82	-	-	-
High EC Control @ 25.07 mS/cm	23420	22.6	23.7	6.7	8.3	7.65	7.73	-	-	-
Grizzly Bay at Dolphin (602)	16585	22.4	23.7	6.9	8.1	7.62	7.84	-	-	-
Suisun Bay off Chipps Island (508)	5855	22.4	23.6	7.3	8.5	7.55	7.92	-	-	-
Montezuma Slough at Nurse Slough (609)	12440	22.4	23.7	7.2	8.5	7.76	7.84	-	-	-
Carquinez Strait, West of Benicia army dock (405)	23025	22.5	23.6	6.9	8.1	7.63	7.76	-	-	-
Suisun Slough at Rush Ranch	13000	22.5	23.9	7.0	8.9	7.53	8.03	-	-	-
Sacramento River at Hood DWR Station	197.2	22.5	23.5	6.9	8.7	7.71	8.30	-	-	-
Rough and Ready DWR station, Stockton	543.5	22.4	23.6	7.3	8.7	7.72	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 B 101-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 10/01/09 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 9/29/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	93	7.5	100	0.0	NS
Low EC Control @ 177.9 µS	98	2.5	98	2.5	NS
Sacramento River Deep Water Channel, Light 55	100	0.0	100	0.0	NS
Upper Cache Slough at mouth of Ulatis Creek	100	0.0	100	0.0	NS
Confluence of Lindsey Sl. and Cache Sl.	100	0.0	100	0.0	NS
Sacramento River at tip of Grand Island (711)[3]	98	2.5	93	2.5	NS
San Joaquin River at Potato Slough (815)	100	0.0	100	0.0	NS
Old River, western arm at railroad bridge (902)	100	0.0	98	2.5	NS
Old River at mouth of Holland Cut (915)	100	0.0	100	0.0	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.049	0.009	0.044	0.004	NS
Low EC Control @ 177.9 µS	0.046	0.007	0.056	0.008	NS
Sacramento River Deep Water Channel, Light 55	0.096	0.006	0.096	0.007	NS
Upper Cache Slough at mouth of Ulatis Creek	0.084	0.013	0.082	0.013	NS
Confluence of Lindsey Sl. and Cache Sl.	0.070	0.012	0.084	0.006	NS
Sacramento River at tip of Grand Island (711)[3]	0.056	0.005	0.068	0.004	NS
San Joaquin River at Potato Slough (815)	0.083	0.010	0.092	0.004	NS
Old River, western arm at railroad bridge (902)	0.093	0.003	0.081	0.011	NS
Old River at mouth of Holland Cut (915)	0.070	0.012	0.088	0.006	NS

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Samples were evaluated using standard USEPA single-concentration statistical procedures.
2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.
3. This low conductivity sample was compared to the Low EC Control.

Table B 101-2. Summary of 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 9/29/09.

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 Deep Water Channel, Light 55	295	21.1	7.15	8.7	22.0	0.06	0.000
Upper Cache Slough at mouth of Ulatis Creek	241	18.7	7.48	9.5	55.9	0.07	0.001
Confluence of Lindsey Sl. and Cache Sl.	219	19.6	7.46	9.0	34.1	0.11	0.001
Sacramento River at tip of Grand Island (711)	174	20.4	7.40	8.8	4.5	0.48	0.005
San Joaquin River at Potato Slough (815)	336	21.7	7.35	8.4	3.2	0.08	0.001
Old River, western arm at railroad bridge (902)	656	22.1	7.45	8.7	4.9	0.03	0.000
Old River at mouth of Holland Cut (915)	598	22.3	7.43	8.4	3.2	0.04	0.000

Table B 101-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 10/01/09 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 9/29/09.

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	22.2	23.7	7.8	8.3	7.73	8.15	102	60	-
Low EC Control @ 177.9 µS	177	22.2	24.0	7.7	8.5	7.49	7.95	52	32	-
Sacramento River Deep Water Channel, Light 55	289	22.3	23.8	7.3	8.8	7.90	8.16	88	86	0.004
Upper Cache Slough at mouth of Ulatis Creek	230	22.1	23.7	7.6	8.4	7.95	8.21	80	91	0.005
Confluence of Lindsey Sl. and Cache Sl.	211	22.6	23.8	7.5	8.3	7.87	8.14	74	83	0.007
Sacramento River at tip of Grand Island (711)	171	22.5	23.9	7.5	8.5	7.67	7.93	60	70	0.019
San Joaquin River at Potato Slough (815)	333	22.5	23.8	7.4	8.6	7.88	8.06	80	78	0.004
Old River, western arm at railroad bridge (902)	612	22.4	23.9	7.5	8.6	7.85	8.10	108	78	0.002
Old River at mouth of Holland Cut (915)	598	22.7	23.7	7.4	8.4	7.87	8.15	106	82	0.002
DIEPAMHR + 25 ppb PBO	335	22.6	23.1	7.3	8.2	7.76	8.14	-	-	-
Low EC Control @ 177.9 µS + 25 ppb PBO	176	22.6	23.3	7.6	8.6	7.49	7.95	-	-	-
Sacramento River Deep Water Channel, Light 55 + 25 ppb PBO	292	22.5	23.4	7.3	8.6	7.93	8.13	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	232	22.6	23.5	7.5	8.4	7.93	8.17	-	-	-
Confluence of Lindsey Sl. and Cache Sl. + 25 ppb PBO	211	22.5	23.6	7.6	8.5	7.88	8.13	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	171	22.5	23.6	7.5	8.3	7.72	8.08	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	331	22.5	23.7	7.4	8.4	7.87	8.14	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	648	22.6	23.8	7.5	8.5	7.80	8.10	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	598	22.6	23.8	7.4	8.3	7.85	8.14	-	-	-

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 B 102-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 10/02/09 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 9/30/09 - 10/01/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	100	0.0	98	2.5	NS
Low EC Control @ 166.3 µS/cm	98	2.3	100	0.0	NS
High EC Control @ 15.63 mS/cm	93	4.8	95	2.9	NS
High EC Control @ 19.62 mS/cm	100	0.0	93	4.8	NS
Montezuma Slough at Nurse Slough (609)[4]	95	5.0	90	7.1	NS
Grizzly Bay at Dolphin (602)[5]	95	3.1	81	7.4	NS
Suisun Bay off Chipps Island (508)[3]	100	0.0	100	0.0	NS
Sacramento River at Hood DWR Station	100	0.0	100	0.0	NS
Rough and Ready DWR station, Stockton	100	0.0	100	0.0	NS
Suisun Slough at Rush Ranch[4]	95	2.6	98	2.5	NS
Bottle Blank: DIEPAMHR	92	5.3	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.042	0.012	0.052	0.004	NS
Low EC Control @ 166.3 µS/cm	0.038	0.001	0.041	0.006	NS
High EC Control @ 15.63 mS/cm	0.036	0.005	0.032*	0.000	NS
High EC Control @ 19.62 mS/cm	0.043	0.006	0.030*	0.002	NS
Montezuma Slough at Nurse Slough (609)[4]	0.064	0.006	0.067	0.004	NS
Grizzly Bay at Dolphin (602)[5]	0.051	0.004	0.054	0.008	NS
Suisun Bay off Chipps Island (508)[3]	0.067	0.009	0.063	0.004	NS
Sacramento River at Hood DWR Station	0.051	0.009	0.077	0.004	S* (151%)
Rough and Ready DWR station, Stockton	0.110	0.002	0.103	0.007	NS
Suisun Slough at Rush Ranch[4]	0.065	0.006	0.075	0.009	NS
Bottle Blank: DIEPAMHR	0.054	0.010	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Samples were evaluated using standard USEPA single-concentration statistical procedures.

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

*: $P < 0.05$

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

4. This high conductivity sample was compared to the High EC Control @ 15.63 mS/cm.

5. This high conductivity sample was compared to the High EC Control @ 19.62 mS/cm.

Table B 102-2. Summary of 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 9/30/09 - 10/01/09.

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)	14120	20.0	7.30	8.2	15.8	0.11	0.001
Grizzly Bay at Dolphin (602)	18240	19.0	7.47	8.8	16.0	0.15	0.001
Suisun Bay off Chipps Island (508)	6640	20.0	7.26	8.8	7.9	0.10	0.001
Sacramento River at Hood DWR Station	161	19.7	7.44	8.0	5.8	0.34	0.003
Rough and Ready DWR station, Stockton	521	22.2	7.28	6.9	8.4	0.09	0.001
Suisun Slough at Rush Ranch	14660	13.3	7.07	7.4	28.6	0.14	0.000
Bottle Blank: DIEPAMHR	-	-	-	-	0.3	0.06	-

Table B 102-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 10/2/09 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 9/30/09-10/1/09.

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 (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	337	22.8	23.6	7.2	8.5	7.72	8.08	102	60	-
Low EC Control @ 166.3 µS	171	22.8	23.7	7.1	8.7	7.48	7.88	52	30	-
High EC Control @ 15.63 mS	15105	22.9	24.0	7.2	8.3	7.68	7.83	1920	78	-
High EC Control @ 19.62 mS	18495	22.7	23.5	7.1	8.4	7.65	7.83	2360	80	-
Montezuma Slough at Nurse Slough (609)	13620	22.6	23.9	7.0	8.4	7.77	7.92	1760	104	0.002
Grizzly Bay at Dolphin (602)	17510	22.7	23.4	6.9	8.7	7.71	7.92	1640	88	0.003
Suisun Bay off Chipps Island (508)	6340	22.7	23.9	7.1	8.8	7.73	7.90	760	80	0.003
Sacramento River at Hood DWR Station	170	22.7	24.3	7.0	8.6	7.39	8.01	60	64	0.017
Rough and Ready Island, DWR Station, Stockton	521	22.7	24.2	7.1	8.9	7.87	8.03	120	86	0.004
Suisun Slough at Rush Ranch	13930	22.7	24.2	7.1	8.5	7.63	8.18	1400	190	0.002
Bottle Blank: DIEPAMHR	345	22.4	24.3	7.2	8.7	7.77	8.06	124	58	0.003
DIEPAMHR + 25 ppb PBO	341	22.5	23.8	7.0	8.2	7.74	8.03	-	-	-
Low EC Control @ 166.3 µS + 25 ppb PBO	175	22.6	23.5	7.3	8.9	7.46	7.82	-	-	-
High EC Control @ 15.63 mS + 25 ppb PBO	15350	22.6	23.4	7.1	8.5	7.61	7.84	-	-	-
High EC Control @ 19.62 mS + 25 ppb PBO	18940	22.6	23.5	6.9	8.2	7.69	7.77	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	13515	22.5	23.4	7.1	8.6	7.76	7.96	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	17280	22.6	23.5	6.9	8.2	7.72	7.89	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	6350	22.5	23.4	7.1	8.7	7.72	7.92	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	192	22.7	23.4	6.7	8.7	7.65	8.06	-	-	-
Rough and Ready Island, DWR Station, Stockton + 25 ppb PBO	525	22.5	23.3	7.4	8.6	7.91	8.09	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	13910	22.6	23.3	6.8	8.6	7.78	8.80	-	-	-

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 B 103-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 10/15/09 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 10/14/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	98	2.5	100	0.0	NS
Low EC Control @ 136.6 µS/cm	100	0.0	100	0.0	NS
High EC Control @ 16.19 mS/cm	100	0.0	100	0.0	NS
High EC Control @ 20.97 mS/cm	98	2.5	95	2.9	NS
Sacramento River at Hood DWR Station[3]	87	6.3	94	3.3	NS
Napa River at River Park Blvd.	100	0.0	97	2.8	NS
Montezuma Slough at Nurse Slough (609)[4]	98	2.5	100	0.0	NS
Suisun Bay off Chipps Island (508)	100	0.0	100	0.0	NS
Grizzly Bay at Dolphin (602)[5]	100	0.0	95	2.9	NS
Bottle Blank: DIEPAMHR	100	0.0	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.037	0.005	0.044	0.003	NS
Low EC Control @ 136.6 µS/cm	0.039	0.002	0.034	0.004	NS
High EC Control @ 16.19 mS/cm	0.035	0.002	0.036	0.004	NS
High EC Control @ 20.97 mS/cm	0.029	0.007	0.032*	0.002	NS
Sacramento River at Hood DWR Station[3]	0.036	0.004	0.027	0.005	NS
Napa River at River Park Blvd.	0.069	0.007	0.055	0.005	NS
Montezuma Slough at Nurse Slough (609)[4]	0.066	0.008	0.070	0.002	NS
Suisun Bay off Chipps Island (508)	0.062	0.007	0.064	0.003	NS
Grizzly Bay at Dolphin (602)[5]	0.044	0.002	0.050	0.004	NS
Bottle Blank: DIEPAMHR	0.047	0.005	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Samples were evaluated using standard USEPA single-concentration statistical procedures.

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

*: $P < 0.05$

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

4. This high conductivity sample was compared to the High EC Control @ 16.19 mS/cm.

5. This high conductivity sample was compared to the High EC Control @ 20.97 mS/cm.

Table B 103-2. Summary of 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 10/14/09.

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 Hood DWR Station	257	16.7	7.42	8.3	7.7	0.38	0.003
Napa River at River Park Blvd.	5320	15.4	7.20	7.9	67.2	0.22	0.001
Montezuma Slough at Nurse Slough (609)	15450	16.5	7.12	9.2	15.4	0.21	0.001
Suisun Bay off Chipps Island (508)	9090	16.5	7.18	9.4	10.4	0.11	0.000
Grizzly Bay at Dolphin (602)	20170	16.4	7.46	9.8	21.5	0.11	0.001

Table B 103-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 10/15/09 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 10/14/09.

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 (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	345	24.0	24.0	7.2	8.1	7.69	8.18	100	60	-
Low EC Control @ 136.6 µS/cm	142	23.1	23.9	7.1	8.7	7.32	8.00	40	26	-
High EC Control @ 16.19 mS/cm	15490	22.9	24.1	7.0	8.4	7.63	7.80	1840	84	-
High EC Control @ 20.97 mS/cm	20320	22.9	23.8	6.9	8.0	7.62	7.78	2400	92	-
Sacramento River at Hood DWR Station	140	23.0	24.0	6.1	8.7	7.43	7.78	52	52	0.011
Napa River at River Park Blvd.	4891	23.0	23.3	6.8	8.3	7.35	7.56	520	48	0.002
Montezuma Slough at Nurse Slough (609)	14965	23.0	23.6	6.8	8.0	7.69	7.81	1760	90	0.004
Suisun Bay off Chipps Island (508)	9615	23.0	23.2	7.0	8.4	7.67	7.86	1040	86	0.002
Grizzly Bay at Dolphin (602)	19340	23.0	24.0	6.9	7.9	7.61	7.81	2280	98	0.002
Bottle Blank: DIEPAMHR	368	22.9	23.6	7.1	8.2	7.77	8.20	112	60	-
DIEPAMHR + 25 ppb PBO	368	22.7	23.0	6.9	8.2	7.63	8.13	-	-	-
Low EC Control @ 136.6 µS/cm + 25 ppb PBO	151	23.1	23.5	7.0	8.9	7.18	7.96	-	-	-
High EC Control @ 16.19 mS/cm + 25 ppb PBO	15630	23.0	23.4	6.8	8.2	7.65	7.83	-	-	-
High EC Control @ 20.97 mS/cm + 25 ppb PBO	19975	23.1	23.1	6.8	8.0	7.60	7.79	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	165	23.1	24.3	6.4	8.6	7.41	7.95	-	-	-
Napa River at River Park Blvd. + 25 ppb PBO	5005	23.1	24.2	7.0	8.2	7.37	7.58	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	14880	23.2	23.8	6.7	7.7	7.64	7.79	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	9840	23.2	24.2	7.1	8.4	7.63	7.92	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	19565	23.3	24.0	7.0	7.9	7.60	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 B 104-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 10/16/09 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 10/15/09 - 10/16/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR [Control for group A]	93	7.5	92	4.8	NS
Low EC Control @ 95.0 µS/cm [A]	93	4.8	77	10.3	NS
DIEPAMHR [Control for group B]	100	0.0	98	2.5	NS
Upper Cache Slough at mouth of Ulatis Creek [A]	98	2.5	10***	6.1	S* (10%)
Confluence of Lindsey Sl. And Cache Sl. [A]	100	0.0	97	2.8	NS
Sacramento River at tip of Grand Island (711)[3] [A]	100	0.0	89	6.1	NS
Old River, western arm at railroad bridge (902) [B]	100	0.0	98	2.5	NS
San Joaquin River at Potato Slough (815)[3] [B]	100	0.0	100	0.0	NS
Sacramento R. Deep Water Channel, Light 55 [A]	100	0.0	100	0.0	NS
Old River at mouth of Holland Cut (915) [B]	100	0.0	98	2.5	NS
Suisun Slough at Rush Ranch [B]	100	0.0	100	0.0	NS
Rough and Ready DWR station, Stockton [B]	100	0.0	86	5.4	NS
Field Dup.: Upper Cache Slough at mouth of Ulatis Creek [A]	95	2.9	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR [Control for group A]	0.038	0.004	0.044	0.002	NS
Low EC Control @ 95.0 µS/cm [A]	0.018**	0.003	0.038	0.004	S** (211%)
DIEPAMHR [Control for group B]	0.049	0.009	0.032	0.007	NS
Upper Cache Slough at mouth of Ulatis Creek [A]	0.039	0.007	0.045	0.005	NS
Confluence of Lindsey Sl. And Cache Sl. [A]	0.083	0.005	0.062	0.004	S* (75%)
Sacramento River at tip of Grand Island (711)[3] [A]	0.061	0.007	0.055	0.007	NS
Old River, western arm at railroad bridge (902) [B]	0.088	0.007	0.049	0.006	S** (56%)
San Joaquin River at Potato Slough (815)[3] [B]	0.066	0.010	0.074	0.014	NS
Sacramento R. Deep Water Channel, Light 55 [A]	0.081	0.003	0.089	0.013	NS
Old River at mouth of Holland Cut (915) [B]	0.080	0.010	0.066	0.008	NS
Suisun Slough at Rush Ranch [B]	0.073	0.009	0.085	0.013	NS
Rough and Ready DWR station, Stockton [B]	0.085	0.020	0.036	0.007	NS
Field Dup.: Upper Cache Slough at mouth of Ulatis Creek [A]	0.053	0.007	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Samples were evaluated using standard USEPA single-concentration statistical procedures.
2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.
*: $P < 0.05$
**: $P < 0.01$
***: $P < 0.001$
3. This low conductivity sample was compared to the Low EC Control.

Table B 104-2. Summary of 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 10/15/09 - 10/16/09.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Upper Cache Slough at mouth of Ulatis Creek	290	16.4	7.26	6.4	163.7	0.22	0.001
Confluence of Lindsey Sl. And Cache Sl.	220	16.8	7.53	9.3	50.7	0.11	0.001
Sacramento River at tip of Grand Island (711)	135	17.4	7.56	9.4	2.9	0.39	0.004
Old River, western arm at railroad bridge (902)	540	17.7	7.47	9.4	2.5	0.04	0.000
San Joaquin River at Potato Slough (815)	170	17.4	7.41	8.9	2.6	0.23	0.002
Sacramento R. Deep Water Channel, Light 55	320	17.1	7.31	9.4	40.7	0.06	0.000
Old River at mouth of Holland Cut (915)	550	17.9	7.40	9.2	3.5	0.02	0.000
Suisun Slough at Rush Ranch	9540	14.8	7.24	6.3	21.7	0.13	0.000
Rough and Ready DWR station, Stockton	522	18.6	7.20	8.5	5.2	0.09	0.000
Field Dup.: Upper Cache Slough at mouth of Ulatis Creek	290	16.4	7.26	6.4	164.0	0.19	0.001

Table B 104-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 10/16/09 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 10/15/09 - 10/16/09.

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 [Control for group A]	339	22.9	23.6	6.9	8.3	7.58	8.74	100	60	-
Low EC Control @ 95.0 µS/cm [A]	111	23.0	23.5	6.8	8.2	7.28	7.81	32	24	-
DIEPAMHR [Control for group B]	336	23.0	23.5	7.3	8.2	7.73	8.74	100	60	-
Upper Cache Slough at mouth of Ulatis Creek [A]	279	23.1	23.8	7.0	8.1	7.61	8.00	108	88	0.004
Confluence of Lindsey Sl. And Cache Sl. [A]	218	23.1	23.6	7.4	8.4	7.79	8.00	80	78	0.004
Sacramento River at tip of Grand Island (711)[3] [A]	137	23.1	23.2	7.0	8.6	7.57	7.83	60	56	0.012
Old River, western arm at railroad bridge (902) [B]	524	23.0	23.7	7.3	8.5	7.76	7.98	96	76	0.002
San Joaquin River at Potato Slough (815)[3] [B]	160	23.1	24.2	7.2	8.6	7.50	7.85	60	58	0.006
Sacramento R. Deep Water Channel, Light 55 [A]	310	23.1	23.9	6.9	8.3	7.77	8.06	88	84	0.003
Old River at mouth of Holland Cut (915) [B]	543	23.1	23.4	7.4	8.6	7.75	8.05	108	82	0.001
Suisun Slough at Rush Ranch [B]	9355	23.1	24.3	7.2	8.3	7.47	8.08	1160	138	0.001
Rough and Ready DWR station, Stockton [B]	526	23.1	24.3	7.2	8.2	7.80	8.12	124	90	0.004
Field Dup.: Upper Cache Slough at mouth of Ulatis Creek [A]	290	23.1	24.2	7.0	8.1	7.70	8.04	108	88	0.005
DIEPAMHR [Control for group A] + 25 ppb PBO	343	23.2	23.9	6.9	8.2	7.69	8.10	-	-	-
Low EC Control @ 95.0 µS/cm [A] + 25 ppb PBO	110	23.2	24.0	6.9	8.2	7.31	7.77	-	-	-
DIEPAMHR [Control for group B] + 25 ppb PBO	339	23.3	23.3	7.2	8.1	7.62	8.77	-	-	-
Upper Cache Slough at mouth of Ulatis Creek [A] + 25 ppb PBO	288	23.3	24.2	7.1	8.7	7.74	7.88	-	-	-
Confluence of Lindsey Sl. And Cache Sl. [A] + 25 ppb PBO	223	23.3	24.1	7.3	8.3	7.82	8.03	-	-	-
Sacramento River at tip of Grand Island (711)[3] [A] + 25 ppb PBO	137	23.3	24.3	7.1	8.3	7.58	7.87	-	-	-
Old River, western arm at railroad bridge (902) [B] + 25 ppb PBO	533	23.3	24.1	7.3	8.0	7.74	8.10	-	-	-
San Joaquin River at Potato Slough (815)[3] [B] + 25 ppb PBO	162	23.3	24.3	7.0	8.3	7.55	8.10	-	-	-
Sacramento R. Deep Water Channel, Light 55 [A] + 25 ppb PBO	316	23.3	24.3	7.0	8.3	7.74	8.22	-	-	-
Old River at mouth of Holland Cut (915) [B] + 25 ppb PBO	550	23.3	24.3	7.2	8.7	7.79	8.17	-	-	-
Suisun Slough at Rush Ranch [B] + 25 ppb PBO	9335	23.4	24.2	7.5	8.3	7.85	8.10	-	-	-
Rough and Ready DWR station, Stockton [B] + 25 ppb PBO	524	23.5	24.1	7.3	8.4	7.80	8.13	-	-	-

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 B 105-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 10/29/09 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 10/27/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	93	4.8	100	0.0	NS
Low EC Control @ 180.9 µS/cm	95	2.9	93	2.5	NS
Upper Cache Slough at mouth of Ulatis Creek	100	0.0	100	0.0	NS
Sacramento R. Deep Water Channel, Light 55	100	0.0	93	4.8	NS
Sacramento River at tip of Grand Island (711)[3]	95	2.9	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)	100	0.0	100	0.0	NS
San Joaquin River at Potato Slough (815)[3]	98	2.5	100	0.0	NS
Confluence of Lindsey Sl. And Cache Sl.[3]	100	0.0	100	0.0	NS
Bottle Blank	98	2.5	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.055	0.008	0.045	0.007	NS
Low EC Control @ 180.9 µS/cm	0.027*	0.007	0.038	0.010	NS
Upper Cache Slough at mouth of Ulatis Creek	0.072	0.004	0.077	0.003	NS
Sacramento R. Deep Water Channel, Light 55	0.070	0.004	0.076	0.007	NS
Sacramento River at tip of Grand Island (711)[3]	0.051	0.013	0.073	0.006	NS
Old River at mouth of Holland Cut (915)	0.061	0.005	0.116	0.011	S** (190%)
Old River, western arm at railroad bridge (902)	0.088	0.007	0.098	0.009	NS
San Joaquin River at Potato Slough (815)[3]	0.067	0.007	0.094	0.009	NS
Confluence of Lindsey Sl. And Cache Sl.[3]	0.100	0.005	0.080	0.011	NS
Bottle Blank	0.060	0.006	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

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

3. These low conductivity samples were compared to the Low EC Control.

Table B 105-2. Summary of 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 10/27/09.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Upper Cache Slough at mouth of Ulatis Creek	487	15.9	7.14	7.8	42.7	0.25	0.001
Sacramento R. Deep Water Channel, Light 55	337	16.8	7.83	9.0	17.8	0.04	0.001
Sacramento River at tip of Grand Island (711)	176	16.6	7.77	9.1	2.6	0.56	0.009
Old River at mouth of Holland Cut (915)	428	17.1	7.41	9.6	2.4	0.05	0.000
Old River, western arm at railroad bridge (902)	534	16.6	7.72	9.7	18.8	0.06	0.001
San Joaquin River at Potato Slough (815)	242	16.5	7.31	9.0	1.9	0.13	0.001
Confluence of Lindsey Sl. And Cache Sl.	214	17.0	7.38	9.3	20.3	0.19	0.001
Bottle Blank: DIEPAMHR	-	-	-	-	0.1	0.03	-

Table B 105-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 10/29/09 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 10/27/09.

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 (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	349	21.3	22.8	7.8	8.7	7.73	8.17	100	52	-
Low EC Control @ 180.9 µS/cm	193	22.7	23.0	7.7	8.7	7.47	7.99	60	30	-
Upper Cache Slough at mouth of Ulatis Creek	534	22.6	22.7	7.4	8.7	8.03	8.20	152	130	0.014
Sacramento R. Deep Water Channel, Light 55	342	22.8	23.2	7.7	8.6	7.82	8.13	104	82	0.002
Sacramento River at tip of Grand Island (711)	196	22.7	23.0	7.6	8.8	7.64	8.12	60	60	0.032
Old River at mouth of Holland Cut (915)	460	22.7	23.9	7.6	8.6	7.70	8.13	92	64	0.003
Old River, western arm at railroad bridge (902)	569	22.7	23.6	7.6	8.7	7.67	8.14	92	64	0.004
San Joaquin River at Potato Slough (815)	234	22.7	23.6	7.5	8.7	7.67	8.10	64	56	0.007
Confluence of Lindsey Sl. And Cache Sl.	234	22.7	23.3	7.4	8.7	7.71	8.18	76	58	0.013
Bottle Blank: DIEPAMHR	378	22.7	23.4	7.7	8.5	7.70	8.12	108	68	0.002
DIEPAMHR + 25 ppb PBO	376	22.6	23.0	7.8	8.7	7.72	8.11	-	-	-
Low EC Control @ 180.9 µS/cm + 25 ppb PBO	212	22.7	23.8	7.6	8.9	7.46	7.96	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	544	22.7	23.5	7.5	8.9	8.04	8.26	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	297	22.6	23.2	7.8	8.9	7.85	8.22	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	182	22.7	23.6	7.7	8.6	7.61	8.04	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	433	22.7	22.9	7.6	8.8	7.68	8.13	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	553	22.8	23.6	7.6	8.9	7.68	8.13	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	238	22.7	23.7	7.6	8.7	7.72	7.85	-	-	-
Confluence of Lindsey Sl. And Cache Sl. + 25 ppb PBO	216	22.5	23.5	7.6	8.4	7.70	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 B 106-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 10/30/09 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 10/28/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	98	2.5	100	0.0	NS
Low EC Control @ 168.6 µS/cm	100	0.0	98	2.5	NS
High EC Control @ 14.71 mS/cm	100	0.0	100	0.0	NS
High EC Control @ 17.19 mS/cm	93	4.8	84	10.8	NS
High EC Control @ 24.53 mS/cm	82	3.1	61*	2.3	S** (74%)
Montezuma Slough at Nurse Slough (609)[4]	100	0.0	100	0.0	NS
Grizzly Bay at Dolphin (602)[5]	100	0.0	95	2.9	NS
Suisun Bay off Chipps Island (508)	98	2.5	100	0.0	NS
Carquinez Strait, West of Benicia army dock (405)[6]	82	4.5	50	10.0	S* (61%)
Suisun Slough at Rush Ranch[4]	100	0.0	100	0.0	NS
Rough and Ready DWR station, Stockton	100	0.0	100	0.0	NS
Napa River at River Park Blvd.[6]	95	3.1	93	4.8	NS
Sacramento River at Hood DWR Station[3]	100	0.0	90	4.1	NS
Field Dup.: Montezuma Slough at Nurse Slough (609)[4]	100	0.0	-	-	NA
Field Dup.: Napa River at River Park Blvd.[6]	98	2.5	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.050	0.007	0.042	0.008	NS
Low EC Control @ 168.6 µS/cm	0.032*	0.006	0.033	0.002	NS
High EC Control @ 14.71 mS/cm	0.043	0.004	0.036	0.006	NS
High EC Control @ 17.19 mS/cm	0.030*	0.005	0.047	0.006	NS
High EC Control @ 24.53 mS/cm	0.036	0.012	0.035	0.007	NS
Montezuma Slough at Nurse Slough (609)[4]	0.060	0.005	0.064	0.005	NS
Grizzly Bay at Dolphin (602)[5]	0.056	0.006	0.061	0.002	NS
Suisun Bay off Chipps Island (508)	0.075	0.007	0.078	0.008	NS
Carquinez Strait, West of Benicia army dock (405)[6]	0.044	0.006	0.022	0.006	NS
Suisun Slough at Rush Ranch[4]	0.077	0.005	0.084	0.008	NS
Rough and Ready DWR station, Stockton	0.102	0.006	0.099	0.007	NS
Napa River at River Park Blvd.[6]	0.035	0.005	0.032	0.004	NS
Sacramento River at Hood DWR Station[3]	0.032	0.005	0.028	0.006	NS
Field Dup.: Montezuma Slough at Nurse Slough (609)[4]	0.078	0.009	-	-	NA
Field Dup.: Napa River at River Park Blvd.[6]	0.034	0.005	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard statistical protocols.

*: $P < 0.05$

**: $P < 0.01$

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

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

4. These high conductivity samples were compared to the High EC Control @ 14.71 mS/cm.

5. This high conductivity sample was compared to the High EC Control @ 17.19 mS/cm.

6. These high conductivity samples were compared to the High EC Control @ 24.53 mS/cm.

Table B 106-2. Summary of 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 10/28/09 - 10/29/09.

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)	13700	15.5	6.86	9.4	18.7	0.25	0.000
Grizzly Bay at Dolphin (602)	16330	14.0	7.16	10.1	66.2	0.20	0.001
Suisun Bay off Chipps Island (508)	4434	14.0	7.42	9.8	48.6	0.16	0.001
Carquinez Strait, West of Benicia army dock (405)	23330	14.8	6.99	9.5	18.6	0.15	0.000
Suisun Slough at Rush Ranch	12440	15.6	7.02	6.9	39.8	0.32	0.001
Rough and Ready DWR station, Stockton	395	15.4	7.33	9.6	4.4	0.00	0.000
Napa River at River Park Blvd.	21920	14.6	7.17	6.1	13.2	0.44	0.001
Sacramento River at Hood DWR Station	161	14.8	7.29	9.0	6.2	0.34	0.002
Field Dup.: Montezuma Slough at Nurse Slough (609)	13700	15.5	6.86	9.4	20.6	0.23	0.000
Field Dup.: Napa River at River Park Blvd.	21920	14.6	7.17	6.1	18.1	0.42	0.001

Table B 106-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 10/30/09 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 10/28/09 - 10/29/09.

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 (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	365	22.2	22.8	7.6	8.7	7.76	8.02	100	52	-
Low EC Control @ 168.6 µS/cm	209	22.1	23.2	7.7	8.9	7.53	7.91	60	30	-
High EC Control @ 14.71 mS/cm	13920	22.2	23.0	7.2	8.8	7.73	7.80	1680	74	-
High EC Control @ 17.19 mS/cm	16320	22.2	23.0	7.4	8.6	7.72	7.84	2000	74	-
High EC Control @ 24.53 mS/cm	23185	22.2	22.7	7.1	8.9	7.77	7.82	2840	84	-
Montezuma Slough at Nurse Slough (609)	13470	22.2	22.6	7.3	8.6	7.74	7.86	1600	90	0.004
Grizzly Bay at Dolphin (602)	15780	22.3	23.4	7.3	8.9	7.74	7.84	1880	80	0.004
Suisun Bay off Chipps Island (508)	4347	22.2	23.2	7.5	8.8	7.73	7.91	540	72	0.005
Carquinez Strait, West of Benicia army dock (405)	22705	22.6	23.1	6.9	8.8	7.65	7.80	2760	90	0.003
Suisun Slough at Rush Ranch	11805	22.6	23.1	7.2	8.5	7.31	8.38	1560	274	0.002
Rough and Ready DWR station, Stockton	435	22.3	23.1	7.5	8.7	7.77	8.19	92	70	0.000
Napa River at River Park Blvd.	20440	22.7	23.4	7.1	8.6	7.65	7.83	2600	108	0.006
Sacramento River at Hood DWR Station	205	21.6	22.2	7.5	8.7	7.71	7.88	56	60	0.008
Field Dup.: Montezuma Slough at Nurse Slough (609)	13205	22.3	22.7	7.4	8.8	7.74	7.84	1640	86	0.005
Field Dup.: Napa River at River Park Blvd.	20800	22.3	22.8	7.1	8.8	7.73	7.87	2560	104	0.007
DIEPAMHR + 25 ppb PBO	358	22.4	22.6	7.8	8.9	7.78	8.12	-	-	-
Low EC Control @ 168.6 µS/cm + 25 ppb PBO	221	22.3	22.4	7.8	8.9	7.46	7.96	-	-	-
High EC Control @ 14.71 mS/cm + 25 ppb PBO	14045	22.4	22.5	7.5	8.8	7.73	7.84	-	-	-
High EC Control @ 17.19 mS/cm + 25 ppb PBO	16930	22.4	22.9	7.4	8.5	7.74	7.86	-	-	-
High EC Control @ 24.53 mS/cm + 25 ppb PBO	23405	22.4	23.4	7.0	8.3	7.71	7.84	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	13360	22.4	23.0	7.3	8.5	7.77	7.86	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	16060	22.4	23.6	7.3	8.4	7.73	7.91	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	4463	22.4	23.5	7.4	8.9	7.78	7.98	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	22675	22.4	23.4	7.0	8.3	7.66	7.90	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	12130	22.5	23.7	7.3	8.5	7.41	8.38	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	499.9	22.5	23.8	7.2	8.9	7.82	8.11	-	-	-
Napa River at River Park Blvd. + 25 pbb PBO	21275	22.5	23.8	7.1	8.9	7.68	7.84	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	202.9	22.5	24.0	7.6	8.7	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 B 107-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 11/11/09 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 11/09/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	98	2.5	98	2.5	NS
San Joaquin River at Potato Slough (815)	100	0.0	100	0.0	NS
Confluence of Lindsey Sl. And Cache Sl.	100	0.0	100	0.0	NS
Old River at mouth of Holland Cut (915)	100	0.0	100	0.0	NS
Upper Cache Slough at mouth of Ulatis Creek	100	0.0	100	0.0	NS
Sacramento R. Deep Water Channel, Light 55	98	2.5	95	2.9	NS
Sacramento River at tip of Grand Island (711)	98	2.5	100	0.0	NS
Old River, western arm at railroad bridge (902)	100	0.0	100	0.0	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.055	0.004	0.049	0.002	NS
San Joaquin River at Potato Slough (815)	0.107	0.014	0.090	0.002	NS
Confluence of Lindsey Sl. And Cache Sl.	0.081	0.009	0.101	0.005	NS
Old River at mouth of Holland Cut (915)	0.117	0.008	0.092	0.018	NS
Upper Cache Slough at mouth of Ulatis Creek	0.108	0.004	0.122	0.009	NS
Sacramento R. Deep Water Channel, Light 55	0.090	0.012	0.085	0.008	NS
Sacramento River at tip of Grand Island (711)	0.087	0.004	0.106	0.010	NS
Old River, western arm at railroad bridge (902)	0.097	0.009	0.110	0.005	NS

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

Table B 107-2. Summary of 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 11/9/09.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (^{o}C)	pH	DO (mg/L)			
San Joaquin River at Potato Slough (815)	305	15.5	8.55	9.6	2.3	0.13	0.011
Confluence of Lindsey Sl. And Cache Sl.	191	14.7	8.59	9.4	9.4	0.28	0.025
Old River at mouth of Holland Cut (915)	433	15.4	8.61	9.8	2.6	0.02	0.002
Upper Cache Slough at mouth of Ulatis Creek	402	14.5	8.38	8.7	25.4	0.18	0.010
Sacramento R. Deep Water Channel, Light 55	215	14.7	8.38	9.2	12.0	0.24	0.014
Sacramento River at tip of Grand Island (711)	206	14.9	8.60	9.4	7.0	0.23	0.021
Old River, western arm at railroad bridge (902)	518	15.1	8.58	10.2	2.0	0.04	0.003

Table B 107-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 11/11/09 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 11/9/09.

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 (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	342	20.1	23.7	7.5	8.5	7.77	8.05	104	64	-
San Joaquin River at Potato Slough (815)	256	20.5	23.9	7.3	8.5	7.73	7.94	104	60	0.005
Confluence of Lindsey Sl. And Cache Sl.	187	20.9	23.7	7.4	8.6	7.65	7.96	84	68	0.010
Old River at mouth of Holland Cut (915)	423	21.1	24.0	7.3	8.5	7.74	7.94	120	64	0.001
Upper Cache Slough at mouth of Ulatis Creek	406	21.5	23.8	7.1	8.3	8.01	8.21	128	110	0.008
Sacramento R. Deep Water Channel, Light 55	209	21.0	24.1	7.2	8.4	7.75	8.03	80	70	0.012
Sacramento River at tip of Grand Island (711)	214	21.8	24.2	7.1	8.7	7.66	8.03	96	64	0.012
Old River, western arm at railroad bridge (902)	528	21.9	23.9	7.2	8.3	7.75	8.02	108	64	0.002
DIEPAMHR + 25 ppb PBO	343	21.4	22.4	7.1	8.4	7.73	8.04	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	257	21.3	22.8	7.2	8.4	7.63	7.94	-	-	-
Confluence of Lindsey Sl. And Cache Sl. + 25 ppb PBO	193	21.9	22.7	7.2	8.4	7.66	7.98	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	429	21.0	23.0	7.3	8.5	7.76	7.97	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	411	22.1	23.0	7.0	8.4	8.02	8.21	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	209	22.4	23.3	7.2	8.4	7.77	7.99	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	207	21.2	23.1	6.9	8.5	7.69	7.95	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	543	22.6	23.5	7.2	8.4	7.73	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 B 108-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 11/12/09 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 11/10/09 and 11/12/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	100	0.0	95	2.9	NS
Low EC @ 181.5 uS/cm	100	0.0	95	2.9	NS
High EC @ 14.98 mS/cm	100	0.0	100	0.0	NS
High EC @ 18.98 mS/cm	98	2.5	90	4.1	NS
High EC @ 24.24 mS/cm	79	10.0	73**	4.3	NS
Grizzly Bay at Dolphin (602)[5]	100	0.0	95	3.1	NS
Suisun Bay off Chipps Island (508)	100	0.0	100	0.0	NS
Carquinez Strait, West of Benicia army dock (405)[6]	75	10.4	61	10.7	NS
Montezuma Slough at Nurse Slough (609)[4]	100	0.0	100	0.0	NS
Suisun Slough at Rush Ranch[4]	98	2.5	100	0.0	NS
Rough and Ready DWR station, Stockton	100	0.0	100	0.0	NS
Napa River at River Park Blvd.[6]	77	6.2	79	10.7	NS
Sacramento River at Hood DWR Station[3]	100	0.0	98	2.5	NS
Field Dup.: Carquinez Strait, West of Benicia army dock (405)[6]	71	8.0	-	-	NA

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.046	0.005	0.043	0.006	NS
Low EC @ 181.5 uS/cm	0.025*	0.003	0.030	0.003	NS
High EC @ 14.98 mS/cm	0.024*	0.006	0.025*	0.008	NS
High EC @ 18.98 mS/cm	0.035	0.004	0.031	0.003	NS
High EC @ 24.24 mS/cm	0.030*	0.005	0.034	0.003	NS
Grizzly Bay at Dolphin (602)[5]	0.035	0.005	0.055	0.006	S*
Suisun Bay off Chipps Island (508)	0.091	0.007	0.098	0.007	NS
Carquinez Strait, West of Benicia army dock (405)[6]	0.032	0.009	0.042	0.007	NS
Montezuma Slough at Nurse Slough (609)[4]	0.042	0.004	0.055	0.005	NS
Suisun Slough at Rush Ranch[4]	0.073	0.012	0.088	0.007	NS
Rough and Ready DWR station, Stockton	0.102	0.009	0.113	0.004	NS
Napa River at River Park Blvd.[6]	0.027	0.007	0.040	0.001	NS
Sacramento River at Hood DWR Station[3]	0.061	0.005	0.072	0.004	NS
Field Dup.: Carquinez Strait, West of Benicia army dock (405)[6]	0.030	0.003	-	-	NA

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard statistical protocols.
*: $P < 0.05$
2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.
3. This low conductivity sample was compared to the Low EC Control.
4. This high conductivity sample was compared to the High EC Control @ 14.98 mS/cm.
5. This high conductivity sample was compared to the High EC Control @ 18.98 mS/cm.

6. This high conductivity sample was compared to the High EC Control @ 24.24 mS/cm.

Table B 108-2. Summary of 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 11/12/09.

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)	17930	15.4	7.38	9.6	12.2	0.09	0.000
Suisun Bay off Chipps Island (508)	9710	15.3	7.58	9.7	16.2	0.12	0.001
Carquinez Strait, West of Benicia army dock (405)	22400	15.3	7.34	9.5	14.4	0.12	0.000
Montezuma Slough at Nurse Slough (609)	14260	15.7	7.40	7.5	11.9	0.34	0.002
Suisun Slough at Rush Ranch	12340	14.1	7.23	6.9	27.1	0.23	0.001
Rough and Ready DWR station, Stockton	495	15.3	7.54	8.9	5.0	0.07	0.001
Napa River at River Park Blvd.	24240	15.4	7.40	7.9	19.9	0.26	0.001
Sacramento River at Hood DWR Station	182	14.7	7.40	9.4	4.8	0.54	0.003
Field Dup.: Carquinez Strait, West of Benicia army dock (405)	22400	15.3	7.34	9.5	14.0	0.15	0.001

Table B 108-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 11/12/09 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 11/10/09 - 11/12/09.

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	353	22.2	23.9	7.8	8.3	7.74	8.07	104	64	-
Low EC @ 181.5 uS/cm	215	22.5	22.9	7.8	8.5	7.55	7.80	64	18	-
High EC @ 14.98 mS/cm	15020	22.6	23.1	7.6	8.1	7.67	7.77	1680	84	-
High EC @ 18.98 mS/cm	19430	22.6	22.8	7.5	8.1	7.69	7.89	2320	78	-
High EC @ 24.24 mS/cm	24110	22.6	23.2	7.2	8.2	7.66	7.81	3000	84	-
Grizzly Bay at Dolphin (602)	17900	22.7	24.1	7.0	8.2	7.66	7.82	1960	86	0.001
Suisun Bay off Chipps Island (508)	9265	22.6	24.1	7.4	8.3	7.71	7.86	1000	74	0.002
Carquinez Strait, West of Benicia army dock (405)	23105	22.6	24.0	7.1	8.3	7.58	7.87	2840	92	0.002
Montezuma Slough at Nurse Slough (609)	14220	22.6	24.0	7.3	8.4	7.36	7.89	1560	98	0.003
Suisun Slough at Rush Ranch	12165	22.6	23.9	7.3	8.2	7.29	8.14	1440	160	0.002
Rough and Ready DWR station, Stockton	498	22.6	23.7	7.7	8.5	7.64	8.05	100	76	0.001
Napa River at River Park Blvd.	23865	22.6	24.0	7.2	8.2	7.34	7.88	2760	116	0.002
Sacramento River at Hood DWR Station	10098	22.6	23.7	7.4	8.9	7.53	7.94	60	66	0.009
Field Dup.: Carquinez Strait, West of Benicia army dock (405)	22695	22.6	24.3	7.2	8.7	7.60	7.83	2360	96	0.002
DIEPAMHR + 25 ppb PBO	378	22.6	24.3	7.8	8.3	7.79	8.01	-	-	-
Low EC @ 181.5 uS/cm + 25 ppb PBO	205	22.4	22.6	7.8	8.4	7.54	7.86	-	-	-
High EC @ 14.98 mS/cm + 25 ppb PBO	7360	22.7	23.1	7.7	8.1	7.72	7.80	-	-	-
High EC @ 18.98 mS/cm + 25 ppb PBO	18960	22.9	23.0	7.1	8.2	7.70	7.80	-	-	-
High EC @ 24.24 mS/cm + 25 ppb PBO	23985	23.0	23.1	6.9	7.9	7.68	7.82	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	17595	22.6	24.0	7.6	8.1	7.67	7.83	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	9240	23.0	23.9	7.4	8.4	7.70	7.84	-	-	-
Carquinez Strait, West of Benicia army dock (405) + 25 ppb PBO	23035	23.5	23.6	7.0	8.0	7.67	7.85	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	79035	22.7	24.0	7.4	8.2	7.33	7.89	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	12140	22.8	22.8	7.4	8.2	7.35	8.14	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	598	22.8	23.0	7.3	8.5	7.71	7.96	-	-	-
Napa River at River Park Blvd. + 25 ppb PBO	23200	23.1	23.6	7.0	7.9	7.36	7.89	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	201.9	22.7	23.3	7.6	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 B 109-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 12/3/09 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 12/1/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	94	3.2	95	5.0	NS
Low EC Control @ 211.7 µS/cm	98	2.5	98	2.5	NS
Old River at mouth of Holland Cut (915)	98	2.5	100	0.0	NS
Upper Cache Slough at mouth of Ulatis Creek	98	2.5	100	0.0	NS
Old River, western arm at railroad bridge (902)	98	2.5	98	2.5	NS
Sacramento River at tip of Grand Island (711)[3]	100	0.0	100	0.0	NS
San Joaquin River at Potato Slough (815)	98	2.5	100	0.0	NS
Confluence of Lindsey Sl. And Cache Sl.[3]	100	0.0	100	0.0	NS
Sacramento R. Deep Water Channel, Light 55	98	2.5	100	0.0	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.037	0.005	0.041	0.003	NS
Low EC Control @ 211.7 µS/cm	0.033	0.003	0.041	0.004	NS
Old River at mouth of Holland Cut (915)	0.078	0.008	0.096	0.004	NS
Upper Cache Slough at mouth of Ulatis Creek	0.083	0.006	0.094	0.006	NS
Old River, western arm at railroad bridge (902)	0.086	0.004	0.099	0.005	NS
Sacramento River at tip of Grand Island (711)[3]	0.075	0.004	0.075	0.005	NS
San Joaquin River at Potato Slough (815)	0.086	0.009	0.085	0.007	NS
Confluence of Lindsey Sl. And Cache Sl.[3]	0.081	0.007	0.065	0.004	NS
Sacramento R. Deep Water Channel, Light 55	0.074	0.006	0.099	0.007	S* (134%)

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

3. These low conductivity samples were compared to the Low EC Control.

Table B 109-2. Summary of 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 12/1/09.

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)	462	11.0	7.63	11.2	11.0	0.06	0.000
Upper Cache Slough at mouth of Ulatis Creek	376	10.2	7.90	11.8	22.6	0.08	0.001
Old River, western arm at railroad bridge (902)	563	11.0	7.61	11.0	9.6	0.08	0.001
Sacramento River at tip of Grand Island (711)	215	10.8	7.60	10.0	5.4	0.36	0.003
San Joaquin River at Potato Slough (815)	318	11.3	7.66	11.2	4.8	0.13	0.001
Confluence of Lindsey Sl. And Cache Sl.	229	10.6	7.61	11.1	18.4	0.29	0.002
Sacramento R. Deep Water Channel, Light 55	281	10.7	7.83	11.2	18.0	0.13	0.002

Table B 109-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 12/3/09 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 12/1/09.

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 (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	345	22.3	23.3	7.8	8.7	7.86	8.04	100	58	-
Low EC Control @ 211.7 µS/cm	227	22.8	23.8	7.5	8.7	7.70	7.96	88	36	-
Old River at mouth of Holland Cut (915)	458	22.8	23.5	7.7	8.7	7.80	8.07	96	68	0.002
Upper Cache Slough at mouth of Ulatis Creek	400	22.7	23.9	7.8	8.7	8.06	8.27	120	110	0.004
Old River, western arm at railroad bridge (902)	554	22.7	23.7	7.6	8.8	7.89	8.03	100	66	0.003
Sacramento River at tip of Grand Island (711)	205	22.6	23.4	7.7	8.7	7.85	8.10	76	76	0.016
San Joaquin River at Potato Slough (815)	320	22.6	24.1	7.4	8.7	7.82	8.04	84	70	0.005
Confluence of Lindsey Sl. And Cache Sl.	226	22.6	24.2	7.6	8.8	7.86	8.13	88	84	0.015
Sacramento R. Deep Water Channel, Light 55	287	22.9	23.8	7.6	8.7	7.97	8.15	94	86	0.007
DIEPAMHR + 25 ppb PBO	343	22.1	22.9	7.5	8.8	7.82	8.06	-	-	-
Low EC Control @ 211.7 µS/cm + 25 ppb PBO	216	22.5	22.9	7.7	8.9	7.74	7.89	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	452	22.4	22.9	7.7	8.9	7.88	8.05	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	387	22.3	22.9	7.8	8.8	8.12	8.25	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	553	22.1	23.0	7.8	8.7	7.88	8.08	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	214	22.2	23.0	7.7	8.9	7.91	8.10	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	318	22.2	23.0	7.7	8.7	7.87	8.03	-	-	-
Confluence of Lindsey Sl. And Cache Sl. + 25 ppb PBO	229	22.1	23.1	7.7	8.9	7.92	8.17	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	279	22.1	23.0	7.6	8.8	8.00	8.14	-	-	-

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 B 110-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 12/04/09 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 12/02/09 - 12/03/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	100	0.0	98	2.5	NS
High EC Control @ 13.22 mS/cm	100	0.0	100	0.0	NS
High EC Control @ 24.40 mS/cm	95	3.1	86*	2.9	NS
Montezuma Slough at Nurse Slough (609)[3]	98	2.5	98	2.5	NS
Grizzly Bay at Dolphin (602)[4]	95	2.9	93	2.5	NS
Suisun Bay off Chipps Island (508)[3]	98	2.5	100	0.0	NS
Rough and Ready DWR station, Stockton	98	2.5	98	2.5	NS
Suisun Slough at Rush Ranch[3]	100	0.0	100	0.0	NS
Sacramento River at Hood DWR Station	100	0.0	100	0.0	NS
Napa River at River Park Blvd.[4]	100	0.0	92	4.8	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.051	0.005	0.050	0.006	NS
High EC Control @ 13.22 mS/cm	0.043	0.003	0.040	0.005	NS
High EC Control @ 24.40 mS/cm	0.036*	0.004	0.035	0.008	NS
Montezuma Slough at Nurse Slough (609)[3]	0.079	0.011	0.087	0.007	NS
Grizzly Bay at Dolphin (602)[4]	0.047	0.005	0.049	0.007	NS
Suisun Bay off Chipps Island (508)[3]	0.070	0.007	0.087	0.006	NS
Rough and Ready DWR station, Stockton	0.127	0.018	0.105	0.010	NS
Suisun Slough at Rush Ranch[3]	0.124	0.012	0.078	0.005	S* (63%)
Sacramento River at Hood DWR Station	0.101	0.004	0.084	0.007	S* (83%)
Napa River at River Park Blvd.[4]	0.045	0.003	0.049	0.005	NS

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

3. These high conductivity samples were compared to the High EC control @ 13.22 mS/cm.

4. These high conductivity samples were compared to the High EC control @ 24.40 mS/cm.

Table B 110-2. Summary of 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 12/2/09 - 12/3/09.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (^{o}C)	pH	DO (mg/L)			
Montezuma Slough at Nurse Slough (609)	13340	10.7	7.68	10.0	25.5	0.36	0.002
Grizzly Bay at Dolphin (602)	23510	11.4	7.81	10.5	15.0	0.18	0.002
Suisun Bay off Chipps Island (508)	12060	11.4	7.84	10.7	8.2	0.16	0.002
Rough and Ready DWR station, Stockton	707	11.9	7.68	10.9	2.6	0.10	0.001
Suisun Slough at Rush Ranch	13000	7.8	7.23	11.3	11.6	0.15	0.000
Sacramento River at Hood DWR Station	205	11.2	7.60	11.0	6.5	0.55	0.004
Napa River at River Park Blvd.	23040	10.2	7.59	11.3	4.2	0.10	0.000

Table B 110-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 12/4/09 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 12/2/09 - 12/3/09.

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 (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	342	23.1	23.5	7.4	8.9	7.85	7.99	100	58	-
High EC Control @ 13.22 mS/cm	13185	23.1	23.2	7.5	8.8	7.70	7.84	1720	72	-
High EC Control @ 24.40 mS/cm	24025	23.2	23.6	7.2	8.3	7.62	7.83	2880	82	-
Montezuma Slough at Nurse Slough (609)	12615	23.1	23.4	7.2	8.8	7.59	7.93	1680	98	0.005
Grizzly Bay at Dolphin (602)	22985	23.2	23.6	7.0	8.5	7.58	7.90	2800	92	0.002
Suisun Bay off Chipps Island (508)	11880	23.2	23.4	7.2	8.5	7.75	7.86	2120	80	0.003
Rough and Ready DWR station, Stockton	700	23.2	23.6	7.4	8.9	7.88	8.15	204	92	0.003
Suisun Slough at Rush Ranch	12570	23.2	24.1	7.1	8.8	7.41	8.17	1880	162	0.001
Sacramento River at Hood DWR Station	206	23.2	23.9	7.3	8.9	7.84	8.02	100	76	0.019
Napa River at River Park Blvd.	22590	23.2	24.1	6.9	8.5	7.56	7.97	2800	120	0.001
DIEPAMHR + 25 ppb PBO	354	23.3	23.4	7.6	8.9	7.86	8.06	-	-	-
High EC Control @ 13.22 mS/cm + 25 ppb PBO	13180	23.2	24.0	7.5	8.9	7.67	7.93	-	-	-
High EC Control @ 24.40 mS/cm + 25 ppb PBO	23780	23.0	23.3	7.1	8.5	7.63	7.88	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	12395	23.2	23.5	7.1	8.6	7.71	7.94	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	23565	23.3	23.3	7.0	8.2	7.59	7.90	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	11855	23.4	23.8	7.3	8.3	7.75	7.89	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	708	23.2	23.4	7.6	8.8	7.93	8.14	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	12735	23.3	23.4	7.2	8.5	7.51	8.17	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	215	23.7	23.9	7.3	8.6	7.78	8.04	-	-	-
Napa River at River Park Blvd. + 25 ppb PBO	22360	23.3	23.5	7.0	8.4	7.63	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 B 111-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 12/17/09 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 12/15/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	92	2.6	100	0.0	NS
Sacramento R. Deep Water Channel, Light 55	98	2.5	94	3.2	NS
Confluence of Lindsey Sl. And Cache Sl.	100	0.0	98	2.5	NS
Upper Cache Slough at mouth of Ulatis Creek	100	0.0	100	0.0	NS
Sacramento River at tip of Grand Island (711)	90	0.4	90	4.1	NS
San Joaquin River at Potato Slough (815)	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)	100	0.0	100	0.0	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.034	0.003	0.030	0.005	NS
Sacramento R. Deep Water Channel, Light 55	0.068	0.004	0.043	0.005	S** (63%)
Confluence of Lindsey Sl. And Cache Sl.	0.065	0.005	0.049	0.006	NS
Upper Cache Slough at mouth of Ulatis Creek	0.083	0.008	0.061	0.007	NS
Sacramento River at tip of Grand Island (711)	0.076	0.011	0.053	0.004	NS
San Joaquin River at Potato Slough (815)	0.074	0.007	0.082	0.006	NS
Old River at mouth of Holland Cut (915)	0.070	0.002	0.061	0.005	NS
Old River, western arm at railroad bridge (902)	0.064	0.014	0.061	0.010	NS

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

**: $P < 0.01$

Table B 111-2. Summary of 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 12/15/09.

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	291	8.9	7.59	11.5	9.6	0.12	0.001
Confluence of Lindsey Sl. And Cache Sl.	235	8.5	7.59	11.6	7.9	0.34	0.002
Upper Cache Slough at mouth of Ulatis Creek	503	8.4	7.91	11.8	10.9	0.06	0.001
Sacramento River at tip of Grand Island (711)	216	8.5	7.60	11.7	6.6	0.41	0.003
San Joaquin River at Potato Slough (815)	363	9.0	7.61	11.7	4.7	0.22	0.001
Old River at mouth of Holland Cut (915)	586	9.1	7.67	11.6	2.7	0.06	0.000
Old River, western arm at railroad bridge (902)	758	9.4	7.70	11.3	2.6	0.09	0.001

Table B 111-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 12/17/09 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 12/15/09.

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	23.2	24.7	7.7	8.3	7.75	8.07	108	58	-
Sacramento R. Deep Water Channel, Light 55	297	23.0	25.1	7.3	8.6	7.85	8.12	92	86	0.008
Confluence of Lindsey Sl. And Cache Sl.	240	22.7	24.8	7.6	8.7	7.83	8.07	84	86	0.020
Upper Cache Slough at mouth of Ulatis Creek	517	22.7	25.0	7.0	8.8	8.09	8.23	152	130	0.005
Sacramento River at tip of Grand Island (711)	219	22.7	24.8	7.6	8.8	7.83	8.08	84	80	0.024
San Joaquin River at Potato Slough (815)	319	22.6	24.6	7.5	8.6	7.82	8.05	88	78	0.012
Old River at mouth of Holland Cut (915)	595	22.6	24.7	7.8	8.9	7.79	8.03	104	70	0.003
Old River, western arm at railroad bridge (902)	780	22.7	24.9	7.2	8.7	7.80	8.09	124	74	0.005
DIEPAMHR + 25 ppb PBO	355	22.7	24.8	7.8	8.4	7.77	8.07	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	298	22.7	24.8	7.2	8.8	7.86	8.11	-	-	-
Confluence of Lindsey Sl. And Cache Sl. + 25 ppb PBO	239	22.7	24.7	7.4	8.6	7.87	8.11	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	517	22.9	25.0	7.6	8.9	8.11	8.28	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	231	23.1	24.5	7.5	8.5	7.84	8.08	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	369	23.4	24.9	7.3	8.8	7.84	8.04	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	583	23.4	24.9	7.6	8.9	7.78	8.07	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	772	23.2	24.9	7.4	8.9	7.72	8.07	-	-	-

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 B 112-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 12/18/09 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 12/16/09 - 12/17/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	92	2.6	87	2.3	NS
High EC Control @ 14.39 mS/cm	97	2.8	89	4.5	NS
High EC Control @ 18.34 mS/cm	100	0.0	80	9.0	NS
High EC Control @ 20.27 mS/cm	84	6.6	82	4.5	NS
Suisun Bay off Chipps Island (508)	100	0.0	100	0.0	NS
Grizzly Bay at Dolphin (602)[5]	90	4.1	92	2.6	NS
Montezuma Slough at Nurse Slough (609)[3]	100	0.0	95	2.9	NS
Napa River at River Park Blvd.[4]	95	2.9	100	0.0	NS
Sacramento River at Hood DWR Station	95	2.9	100	0.0	NS
Rough and Ready DWR station, Stockton	100	0.0	98	2.5	NS
Suisun Slough at Rush Ranch	100	0.0	95	2.9	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.045	0.006	0.037	0.008	NS
High EC Control @ 14.39 mS/cm	0.031	0.005	0.024	0.003	NS
High EC Control @ 18.34 mS/cm	0.033	0.004	0.023	0.007	NS
High EC Control @ 20.27 mS/cm	0.019**	0.003	0.024	0.006	NS
Suisun Bay off Chipps Island (508)	0.048	0.005	0.045	0.004	NS
Grizzly Bay at Dolphin (602)[5]	0.040	0.006	0.046	0.003	NS
Montezuma Slough at Nurse Slough (609)[3]	0.061	0.009	0.047	0.005	NS
Napa River at River Park Blvd.[4]	0.042	0.006	0.046	0.002	NS
Sacramento River at Hood DWR Station	0.092	0.011	0.084	0.012	NS
Rough and Ready DWR station, Stockton	0.065	0.002	0.071	0.004	NS
Suisun Slough at Rush Ranch	0.062	0.002	0.076	0.001	S** (123%)

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Samples were evaluated using standard USEPA single-concentration statistical procedures.
2. NS: Nonsignificant, S: Significant (% non-PBO mean), NA: Not applicable.
*: $P < 0.05$
**: $P < 0.01$
3. This low conductivity sample was compared to the High EC Control @ 14.39 mS/cm.
4. This high conductivity sample was compared to the High EC Control @ 18.34 mS/cm.
5. This high conductivity sample was compared to the High EC Control @ 20.27 mS/cm.

Table B 112-2. Summary of 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 12/16/09 - 12/17/09.

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)	7980	9.8	7.81	11.0	11.7	0.20	0.002
Grizzly Bay at Dolphin (602)	20040	9.9	7.79	10.0	9.1	0.16	0.001
Montezuma Slough at Nurse Slough (609)	14010	9.3	7.59		29.7	0.31	0.002
Napa River at River Park Blvd.	17830	9.7	7.36	9.9	5.3	0.13	0.000
Sacramento River at Hood DWR Station	220	10.8	7.67	10.8	17.9	0.39	0.003
Rough and Ready DWR station, Stockton	859	10.4	7.65	10.8	2.4	0.21	0.002
Suisun Slough at Rush Ranch	11590	9.2	7.22	9.4	15.1	0.09	0.000

Table B 112-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 12/18/09 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 12/16/09 - 12/17/09.

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 (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	342	21.7	23.2	7.8	8.5	7.84	8.01	108	58	-
High EC Control @ 14.39 mS/cm	14520	22.3	24.5	7.5	8.4	7.76	8.04	1760	76	-
High EC Control @ 18.34 mS/cm	18655	22.7	24.0	7.6	8.3	7.73	7.92	2200	86	-
High EC Control @ 20.27 mS/cm	20255	22.6	24.1	7.6	8.3	7.78	7.94	2520	62	-
Suisun Bay off Chipps Island (508)	8560	22.6	24.1	7.7	8.5	7.78	7.98	888	88	0.005
Grizzly Bay at Dolphin (602)	19265	22.7	24.7	7.5	8.6	7.70	7.95	2320	98	0.003
Montezuma Slough at Nurse Slough (609)	13890	22.6	24.4	7.5	8.5	7.67	7.97	1760	102	0.005
Napa River at River Park Blvd.	17315	22.3	24.7	7.5	8.9	7.62	8.04	2120	126	0.002
Sacramento River at Hood DWR Station	239	22.5	24.6	7.8	8.4	7.90	8.12	84	90	0.016
Rough and Ready DWR station, Stockton	6262	22.3	24.9	7.8	8.7	7.92	8.19	172	100	0.008
Suisun Slough at Rush Ranch	6042	22.4	24.6	7.6	8.2	7.59	8.25	1480	180	0.001
DIEPAMHR + 25 ppb PBO	618	22.3	24.2	8.0	8.5	7.78	8.03	-	-	-
High EC Control @ 14.39 mS/cm + 25 ppb PBO	14085	22.4	24.6	7.2	8.6	7.72	8.08	-	-	-
High EC Control @ 18.34 mS/cm + 25 ppb PBO	18780	22.2	24.7	7.6	8.4	7.77	7.95	-	-	-
High EC Control @ 20.27 mS/cm + 25 ppb PBO	19975	22.3	24.6	7.4	8.5	7.77	7.94	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	8495	22.2	24.2	7.6	8.4	7.84	7.98	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	19420	22.2	24.5	7.5	8.6	7.79	7.93	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	13695	22.4	24.3	7.3	8.5	7.79	7.98	-	-	-
Napa River at River Park Blvd. + 25 ppb PBO	17245	22.1	24.8	7.4	8.5	7.70	8.05	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	259	22.4	24.4	7.6	8.6	7.91	8.12	-	-	-
Rough and Ready DWR station, Stockton + 25 ppb PBO	899	22.3	24.9	7.5	8.6	7.98	8.20	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	11520	22.3	24.4	7.3	8.6	7.70	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 B 113-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 12/30/09 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 12/28/09 - 12/29/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	100	0.0	98	2.5	NS
High E.C. Control @ 12.52 mS/cm	100	0.0	100	0.0	NS
High E.C. Control @ 20.15 mS/cm	97	2.8	98	2.5	NS
High E.C. Control @ 25.23 mS/cm	100	0.0	90	7.1	NS
Suisun Slough at Rush Ranch[3]	100	0.0	98	2.5	NS
Rough and Ready Island, DWR Station, Stockton	100	0.0	100	0.0	NS
Sacramento River at Hood DWR Station	100	0.0	100	0.0	NS
Napa River at River Park Blvd.[4]	100	0.0	100	0.0	NS
Grizzly Bay at Dolphin (602)[5]	100	0.0	90	4.1	NS
Montezuma Slough at Nurse Slough (609)	100	0.0	100	0.0	NS
Suisun Bay off Chipps Island (508)[3]	100	0.0	100	0.0	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.054	0.005	0.052	0.003	NS
High E.C. Control @ 12.52 mS/cm	0.029**	0.003	0.034*	0.004	NS
High E.C. Control @ 20.15 mS/cm	0.022**	0.005	0.036*	0.005	NS
High E.C. Control @ 25.23 mS/cm	0.025*	0.007	0.030*	0.006	NS
Suisun Slough at Rush Ranch[3]	0.095	0.005	0.092	0.003	NS
Rough and Ready Island, DWR Station, Stockton	0.087	0.005	0.078	0.007	NS
Sacramento River at Hood DWR Station	0.101	0.004	0.103	0.005	NS
Napa River at River Park Blvd.[4]	0.062	0.015	0.069	0.007	NS
Grizzly Bay at Dolphin (602)[5]	0.051	0.006	0.038	0.005	NS
Montezuma Slough at Nurse Slough (609)	0.089	0.007	0.086	0.008	NS
Suisun Bay off Chipps Island (508)[3]	0.078	0.006	0.080	0.005	NS

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. These high conductivity samples were compared to the High EC Control @ 12.52 mS/cm.
4. These high conductivity samples were compared to the High EC Control @ 20.15 mS/cm.
5. These high conductivity samples were compared to the High EC Control @ 25.23 mS/cm.

*: $P < 0.05$

**: $P < 0.01$

Table B 113-2. Summary of 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 12/28/09 - 12/29/09.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (°C)	pH	DO (mg/L)			
Suisun Slough at Rush Ranch	11980	8.4	7.28	9.0	16.7	0.12	0.000
Rough and Ready Island, DWR Station, Stockton	831	9.5	7.56	10.7	2.3	0.15	0.001
Sacramento River at Hood DWR Station	214	8.9	7.37	11.2	8.1	0.44	0.002
Napa River at River Park Blvd.	19120	9.9	7.64	11.4	27.5	0.08	0.000
Grizzly Bay at Dolphin (602)	23550	9.5	7.65	11.4	12.1	0.19	0.001
Montezuma Slough at Nurse Slough (609)	8670	9.2	7.22	11.2	19.5	0.28	0.001
Suisun Bay off Chipps Island (508)	11880	9.0	7.36	11.8	7.8	0.24	0.001

Table B 113-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 12/30/09 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 12/28/09 - 12/29/09.

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	21.9	23.6	7.5	8.4	7.73	8.12	104	54	-
High E.C. Control @ 12.52 mS/cm	12025	21.5	23.7	7.4	8.4	7.67	7.83	1600	78	-
High E.C. Control @ 20.15 mS/cm	19445	21.1	23.9	7.3	8.0	7.69	7.80	2640	88	-
High E.C. Control @ 25.23 mS/cm	24550	21.2	23.7	7.1	8.1	7.72	7.82	3320	94	-
Suisun Slough at Rush Ranch	11625	21.9	23.7	7.4	8.2	7.77	8.09	1560	166	0.003
Rough and Ready Island, DWR Station, Stockton	894	22.5	23.9	7.7	8.6	7.95	8.15	192	112	0.009
Sacramento River at Hood DWR Station	250	22.6	24.1	6.9	8.5	7.78	8.04	96	88	0.023
Napa River at River Park Blvd.	18915	22.6	23.9	6.9	8.2	7.79	7.90	2480	132	0.002
Grizzly Bay at Dolphin (602)	23445	22.7	23.6	7.1	8.2	7.76	7.88	3040	106	0.004
Montezuma Slough at Nurse Slough (609)	8470	22.6	23.9	7.1	8.2	7.80	7.92	1200	102	0.007
Suisun Bay off Chipps Island (508)	11650	22.6	23.9	7.3	8.5	7.77	7.88	1480	98	0.006
DIEPAMHR + 25 ppb PBO	437	22.6	23.1	7.7	8.2	7.74	8.13	-	-	-
High E.C. Control @ 12.52 mS/cm + 25 ppb PBO	12450	22.6	23.1	7.3	8.4	7.67	7.85	-	-	-
High E.C. Control @ 20.15 mS/cm + 25 ppb PBO	20015	22.6	23.5	7.1	8.1	7.72	7.81	-	-	-
High E.C. Control @ 25.23 mS/cm + 25 ppb PBO	25395	22.6	23.4	7.1	8.2	7.75	7.80	-	-	-
Suisun Slough at Rush Ranch + 25 ppb PBO	11850	22.6	23.9	7.2	8.2	7.85	8.07	-	-	-
Rough and Ready Island, DWR Station, Stockton + 25 ppb PBO	900	22.6	23.8	7.6	8.8	7.92	8.79	-	-	-
Sacramento River at Hood DWR Station + 25 ppb PBO	270	22.5	23.9	7.5	8.5	7.79	8.10	-	-	-
Napa River at River Park Blvd. + 25 ppb PBO	18910	22.5	23.7	7.0	8.2	7.85	7.89	-	-	-
Grizzly Bay at Dolphin (602) + 25 ppb PBO	23600	22.5	24.1	7.1	8.2	7.76	7.83	-	-	-
Montezuma Slough at Nurse Slough (609) + 25 ppb PBO	8565	22.5	23.9	7.3	8.3	7.82	7.91	-	-	-
Suisun Bay off Chipps Island (508) + 25 ppb PBO	11710	22.6	23.8	7.2	8.4	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 B 114-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 12/31/09 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 12/30/09.

Treatment	Survival (%)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	95	2.9	98	2.5	NS
Upper Cache Slough at mouth of Ulatis Creek	100	0.0	98	2.5	NS
Sacramento R. Deep Water Channel, Light 55	100	0.0	100	0.0	NS
Confluence of Lindsey Sl. And Cache Sl.	100	0.0	98	2.5	NS
Old River, western arm at railroad bridge (902)	100	0.0	100	0.0	NS
Sacramento River at tip of Grand Island (711)	98	2.5	100	0.0	NS
San Joaquin River at Potato Slough (815)	100	0.0	100	0.0	NS
Old River at mouth of Holland Cut (915)	95	2.9	100	0.0	NS

Treatment	Weight (mg/surviving individual)[1]				
	Unmanipulated		25 ppb PBO added		
	mean	se	mean	se	vs Non-PBO[2]
DIEPAMHR	0.051	0.004	0.088	0.016	NS
Upper Cache Slough at mouth of Ulatis Creek	0.117	0.011	0.113	0.005	NS
Sacramento R. Deep Water Channel, Light 55	0.113	0.006	0.116	0.006	NS
Confluence of Lindsey Sl. And Cache Sl.	0.096	0.010	0.127	0.008	S* (132%)
Old River, western arm at railroad bridge (902)	0.124	0.005	0.122	0.013	NS
Sacramento River at tip of Grand Island (711)	0.117	0.005	0.145	0.011	NS
San Joaquin River at Potato Slough (815)	0.135	0.015	0.129	0.013	NS
Old River at mouth of Holland Cut (915)	0.131	0.008	0.126	0.008	NS

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

Table B 114-2. Summary of 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 12/30/09.

Treatment	Field Chemistry				Turbidity (NTU)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	SC (uS/cm)	Temp (^{o}C)	pH	DO (mg/L)			
Upper Cache Slough at mouth of Ulatis Creek	565	8.1	7.34	12.1	11.5	0.02	0.000
Sacramento R. Deep Water Channel, Light 55	324	8.9	7.10	11.0	11.6	0.13	0.000
Confluence of Lindsey Sl. And Cache Sl.	293	8.6	7.42	11.9	7.6	0.24	0.001
Old River, western arm at railroad bridge (902)	854	8.7	7.30	12.1	3.3	0.10	0.000
Sacramento River at tip of Grand Island (711)	220	8.4	7.21	12.1	6.5	0.43	0.001
San Joaquin River at Potato Slough (815)	446	8.9	7.26	11.7	4.0	0.18	0.001
Old River at mouth of Holland Cut (915)	719	8.7	7.32	12.2	2.4	0.09	0.000

Table B 114-3. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 12/31/09 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 12/30/09.

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 (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH			
DIEPAMHR	351	22.6	24.5	7.7	8.3	7.75	7.99	104	54	-
Upper Cache Slough at mouth of Ulatis Creek	559	22.8	24.6	7.6	8.5	8.12	8.27	180	158	0.001
Sacramento R. Deep Water Channel, Light 55	333	22.8	24.7	7.7	8.6	7.97	8.14	112	112	0.009
Confluence of Lindsey Sl. And Cache Sl.	291	22.8	24.8	7.7	8.3	7.95	8.13	116	110	0.014
Old River, western arm at railroad bridge (902)	872	22.8	24.8	7.7	8.4	7.88	8.07	156	88	0.005
Sacramento River at tip of Grand Island (711)	215	22.8	24.8	7.6	8.4	7.83	8.11	88	92	0.020
San Joaquin River at Potato Slough (815)	476	22.8	24.9	7.6	8.5	7.86	8.08	108	92	0.010
Old River at mouth of Holland Cut (915)	719	22.8	24.8	7.7	8.5	7.85	8.01	152	86	0.004
DIEPAMHR + 25 ppb PBO	346	22.8	24.1	7.7	8.3	7.71	8.00	-	-	-
Upper Cache Slough at mouth of Ulatis Creek + 25 ppb PBO	551	22.8	24.0	7.7	8.4	8.14	8.29	-	-	-
Sacramento R. Deep Water Channel, Light 55 + 25 ppb PBO	329	22.8	24.4	7.5	8.3	7.98	8.15	-	-	-
Confluence of Lindsey Sl. And Cache Sl. + 25 ppb PBO	287	22.8	23.8	7.7	8.3	7.95	8.14	-	-	-
Old River, western arm at railroad bridge (902) + 25 ppb PBO	869	22.9	24.0	7.7	8.3	7.85	8.01	-	-	-
Sacramento River at tip of Grand Island (711) + 25 ppb PBO	215	22.9	24.0	7.6	8.5	7.85	8.06	-	-	-
San Joaquin River at Potato Slough (815) + 25 ppb PBO	480	23.3	24.0	7.7	8.3	7.83	8.02	-	-	-
Old River at mouth of Holland Cut (915) + 25 ppb PBO	728	23.1	24.0	7.7	8.5	7.88	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.

Appendix C

Summary Tables:

H. transpacificus Ambient Toxicity Tests

Table C 1-1. Results of a *H. transpacificus* (Delta Smelt) 7-day test initiated 3/27/08 evaluating the toxicity of Sacramento River and Delta water samples collected on 3/25/08 - 3/27/08.[1]

Treatment	Survival (%)[2]	
	mean	se
Low EC Control	48.7	5.8
Hatchery Rearing Control	58.3	6.8
High EC Control	82.6	5.6
Low Turbidity Control	56.3	4.4
Suisun Bay at Public Suisun Dock[4]	91.8	3.4
Sacramento River at Hood DWR Station[3]	25.0	11.9
Sacramento R. Deep Water Channel, Light 55[3]	91.7	8.3
Confluence of Lindsey Sl. And Cache Sl.[3]	59.3	6.9
Rough and Ready DWR station, Stockton[3]	42.4	11.0
Napa River at Vallejo Seawall (340)[5]	89.7	4.8

Tukey's MSD: 35.7%

Tukey's PMSD (% of Hatchery Control): 61.1%

1. Smelt used in this experiment were 28 days old at test initiation.

2. Highlighted areas indicate significant reductions in survival compared to the EC-specific control. Data were analyzed using both USEPA standard single concentration statistical protocols and ANOVA with Tukey's multiple comparison procedure.

3. These low conductivity samples were compared to the Low EC control.

4. This intermediate conductivity sample was compared to the hatchery rearing water control.

5. This high conductivity sample was compared to the High EC control.

Table C 1-2. Chemistry of sample waters examined in a *H. transpacificus* (Delta Smelt) 7-day test initiated 3/27/09, evaluating the toxicity of Sacramento River and Delta water samples collected 3/25/08-3/27/08.

Sample	Collection Date and Time	Field Chemistry				
		SC (uS/cm)	EC (uS/cm)	Temp (°C)	DO (mg/L)	pH
Suisun Bay at Public Suisun Dock	3/26/2008 10:15	3759	2842	12.8	8.6	7.97
Sacramento River at Hood DWR Station	3/25/2008 14:00	1045	880	17.1	9.2	7.91
Sacramento R. Deep Water Channel, Light 55	3/26/2008 10:00	364	290	14.8	10.2	8.18
Confluence of Lindsey Sl. And Cache Sl.	3/26/2008 11:05	459	368	15.1	10.1	8.25
Rough and Ready DWR station, Stockton	3/25/2008 10:30	207	173	16.8	8.9	8.10
Napa River at Vallejo Seawall (340)	3/27/2008 10:30	18580	14084	12.9	10.6	8.03

Table C 1-3. Chemistry of sample waters examined in a *H. transpacificus* (Delta Smelt) 7-day test initiated 3/27/08 evaluating the toxicity of Sacramento River and Delta water samples collected on 3/25/08 - 3/27/08.

Treatment	Temp (°C)			EC (uS/cm)			DO (mg/L)		
	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*
Low EC Control	16.3	0.9	8	226	43	8	9.3	0.4	8
Hatchery Rearing Control	16.4	0.7	8	2149	48	8	9.4	0.4	8
High EC Control	16.4	0.8	8	3793	68	8	9.6	0.5	8
Low Turbidity Control	16.5	0.7	8	2199	54	8	9.7	0.3	8
Suisun Bay at Public Suisun Dock	16.3	0.8	8	2982	47	8	9.7	0.2	8
Sacramento River at Hood DWR Station	16.4	0.7	8	199	54	8	9.9	0.2	8
Sacramento R. Deep Water Channel, Light 55	16.3	0.8	8	323	44	8	10.0	0.2	8
Confluence of Lindsey Sl. And Cache Sl.	16.4	0.6	8	335	57	8	9.9	0.2	8
Rough and Ready DWR station, Stockton	16.4	0.7	8	735	32	8	10.0	0.2	8
Napa River at Vallejo Seawall (340)	16.5	0.7	8	14389	248	8	9.7	0.3	8

Treatment	pH			Turbidity (NTU)			Ammonia Nitrogen (mg/L)		
	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*
Low EC Control	7.68	0.15	8	4.9	1.5	4	0.24	0.24	3
Hatchery Rearing Control	7.91	0.10	8	4.0	1.4	4	0.19	0.11	3
High EC Control	7.93	0.11	8	4.9	1.7	4	0.16	0.06	3
Low Turbidity Control	7.93	0.09	8	4.7	1.9	4	0.17	0.14	3
Suisun Bay at Public Suisun Dock	7.91	0.11	8	16.1	1.4	4	0.15	0.06	4
Sacramento River at Hood DWR Station	7.98	0.06	8	2.3	0.8	4	0.38	0.05	4
Sacramento R. Deep Water Channel, Light 55	8.15	0.07	8	13.1	2.4	4	0.21	0.02	4
Confluence of Lindsey Sl. And Cache Sl.	8.12	0.04	8	11.1	1.6	4	0.24	0.02	4
Rough and Ready DWR station, Stockton	8.11	0.08	8	2.6	0.9	4	0.14	0.05	4
Napa River at Vallejo Seawall (340)	7.92	0.07	8	5.1	2.0	4	0.08	0.01	4

Treatment	Un-ionized Ammonia (mg/L)			Hardness (mg/L as $CaCO_3$)	Alkalinity (mg/L as $CaCO_3$)
	Mean	SD	*N*		
Low EC Control	0.003	0.002	3	-	-
Hatchery Rearing Control	0.004	0.002	3	-	-
High EC Control	0.004	0.001	3	-	-
Low Turbidity Control	0.004	0.0025	3	-	-
Suisun Bay at Public Suisun Dock	0.003	0.002	4	528	184
Sacramento River at Hood DWR Station	0.010	0.001	4	84	78
Sacramento R. Deep Water Channel, Light 55	0.008	0.003	4	132	124
Confluence of Lindsey Sl. And Cache Sl.	0.009	0.001	4	128	126
Rough and Ready DWR station, Stockton	0.005	0.003	4	200	126
Napa River at Vallejo Seawall (340)	0.001	0.000	4	4880	108

Table C 2-1. Results of a *H. transpacificus* (Delta Smelt) 7-day test initiated 4/10/08 evaluating the toxicity of Sacramento River and Delta water samples collected 4/8/08 - 4/10/08.[1]

Treatment	Survival (%)[2]	
	mean	se
Low EC Control	60.0	4.7
Suisun Bay at Public Suisun Dock[4]	87.5	8.0
Sacramento River at Hood DWR Station[5]	34.5	13.3
Sacramento R. Deep Water Channel, Light 55[3]	72.9	9.2
Confluence of Lindsey Sl. And Cache Sl.[3]	67.8	9.6
Rough and Ready DWR station, Stockton[5]	47.9	9.8
Mid EC Control	72.9	6.3
High EC Control	77.3	5.8
Napa River at Vallejo Seawall (340)[4]	95.8	2.4
Low Turbidity Control	27.1	8.6

1. Smelt used in this experiment were 38 days old at test initiation.

2. Highlighted areas indicate significant reductions in survival compared to the appropriate EC-specific control. Data were analyzed using both USEPA standard single concentration statistical protocols and ANOVA with Tukey's multiple comparison procedure.

3. These low conductivity samples were compared to the Low EC control.

4. This high conductivity sample was compared to the High EC control.

5. This low turbidity sample was compared to the low turbidity control.

	MSD	**PMSD†**
Survival (%)	**40.1**	**55.0**

†. Calculated as % of Hatchery Water control

Table C 2-2. Chemistry of sample waters examined in a *H. transpacificus* (Delta Smelt) 7-day test initiated 4/10/08, evaluating the toxicity of Sacramento River and Delta water samples collected on 4/08/08 - 4/10/08.

Sample	Collection Date and Time	Field Chemistry				
		SC (uS/cm)	EC (uS/cm)	Temp (°C)	DO (mg/L)	pH
Suisun Bay at Public Suisun Dock	4/9/2008 11:30	4603	3636	14.5	8.6	7.15
Sacramento River at Hood DWR Station	4/8/2008 14:30	221.3	184	16.6	9.3	8.05
Sacramento R. Deep Water Channel, Light 55	4/9/2008 10:50	395.5	312	14.4	10.6	8.24
Confluence of Lindsey Sl. And Cache Sl.	4/9/2008 11:50	358.8	283	14.4	10.5	8.23
Rough and Ready DWR station, Stockton	4/8/2008 11:00	716	589	16.1	9.8	7.97
Napa River at Vallejo Seawall (340)	4/10/2008 12:15	23250	18321	14.4	10.0	7.92

Sample	Turbidity (NTU)	Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)[1]
Suisun Bay at Public Suisun Dock	84.6	0.04	0.000
Sacramento River at Hood DWR Station	8.1	0.59	0.019
Sacramento R. Deep Water Channel, Light 55	28.9	0.15	0.006
Confluence of Lindsey Sl. And Cache Sl.	29.4	0.21	0.008
Rough and Ready DWR station, Stockton	11.1	0.07	0.002
Napa River at Vallejo Seawall (340)	22.4	0.00	0.000

1. This unionized ammonia measurement was calculated from ammonia nitrogen measured at sample receipt and water chemistry measured at sample collection.

Table C 2-3. Chemistry of sample waters examined in a *H. transpacificus* (Delta Smelt) 7-day test initiated 4/10/08 evaluating the toxicity of Sacramento River and Delta water samples collected 4/8/08 - 4/10/08.

Treatment	Temp (°C)			EC (uS/cm)			DO (mg/L)		
	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*
Low EC Control	16.8	0.4	8	241	60	8	9.2	0.5	8
Suisun Bay at Public Suisun Dock	16.4	0.7	8	3310	101	8	9.5	0.3	8
Sacramento River at Hood DWR Station	16.2	0.8	8	214	63	8	9.6	0.5	8
Sacramento R. Deep Water Channel, Light 55	16.1	0.7	8	334	63	8	9.8	0.3	8
Confluence of Lindsey Sl. And Cache Sl.	16.3	0.7	8	305	37	8	9.8	0.1	8
Rough and Ready DWR station, Stockton	16.1	0.7	8	593	46	8	9.8	0.3	8
Mid EC Control	16.5	0.5	8	639	54	8	9.3	0.3	8
High EC Control	16.5	0.4	8	3281	102	8	9.3	0.3	8
Napa River at Vallejo Seawall (340)	16.4	0.6	8	16830	1565	8	9.6	0.2	8
Low Turbidity Control	16.6	0.4	8	666	49	8	9.6	0.3	8

Treatment	pH			Turbidity (NTU)			Un-ionized Ammonia (mg/L)		
	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*
Low EC Control	7.62	0.12	8	5.8	1.4	6	0.005	0.005	3
Suisun Bay at Public Suisun Dock	7.85	0.20	8	27.0	25.5	7	0.001	0.001	4
Sacramento River at Hood DWR Station	7.85	0.11	8	3.6	2.2	7	0.009	0.003	4
Sacramento R. Deep Water Channel, Light 55	8.02	0.07	8	11.6	7.4	7	0.005	0.003	4
Confluence of Lindsey Sl. And Cache Sl.	8.11	0.07	8	12.2	7.7	7	0.008	0.003	4
Rough and Ready DWR station, Stockton	7.97	0.09	8	4.3	3.1	7	0.003	0.002	4
Mid EC Control	7.82	0.08	8	5.5	1.4	6	0.005	0.004	3
High EC Control	7.78	0.12	8	4.2	1.1	6	0.003	0.003	3
Napa River at Vallejo Seawall (340)	7.81	0.09	8	9.6	6.1	7	0.000	0.000	4
Low Turbidity Control	7.76	0.13	8	2.3	2.2	7	0.001	0.0013	3

Treatment	Hardness (mg/L as $CaCO_3$)	Alkalinity (mg/L as $CaCO_3$)
Low EC Control	-	-
Suisun Bay at Public Suisun Dock	564	168
Sacramento River at Hood DWR Station	84	98
Sacramento R. Deep Water Channel, Light 55	124	119
Confluence of Lindsey Sl. And Cache Sl.	120	117
Rough and Ready DWR station, Stockton	156	96
Mid EC Control	-	-
High EC Control	-	-
Napa River at Vallejo Seawall (340)	2640	112
Low Turbidity Control	-	-

Table C 3-1. Results of a *H. transpacificus* (Delta Smelt) 7-day test initiated 4/24/08 evaluating the toxicity of Sacramento River and Delta water samples collected 4/22/08 - 4/24/08.[1]

Treatment	Survival (%)[1]	
	mean	se
Low EC Control	37.7	3.9
Suisun at Rush Ranch[5]	97.7	2.3
Sacramento River at Hood DWR Station[3,6,7]	20.2	4.9
Sacramento River, Deep Water Channel, Light 55[3]	52.1	9.5
Confluence of Lindsey Slough and Cache Slough (CL)[3]	61.9	7.1
Rough and Ready DWR Station, Stockton[4]	51.3	6.6
Mid EC Control	38.3	15.4
High EC Control	60.8	14.8
Napa River at Vallejo Seawall (340)[5]	76.1	2.0
Low Turbidity Control	50.0	11.4

1. Smelt used in this experiment were 37 days old at test initiation.

2. Highlighted areas indicate significant reductions in survival compared to the appropriate EC-specific and turbidity-specific controls. Data were analyzed using both USEPA standard single concentration statistical protocols and ANOVA with Tukey's multiple comparison procedure.

3. These low conductivity samples were compared to the Low EC control.

4. This intermediate conductivity sample was compared to the Mid EC control.

5. This high conductivity sample was compared to the High EC control.

6. This low turbidity sample was compared to the low turbidity control.

7. Hood showed lower survival than both the low EC control and the low turbidity control according to USEPA statistical protocols, but not when examined with Tukey's multiple comparison procedure.

	MSD	**PMSD†**
Survival (%)	**43.7**	**114.2**

†. Calculated as % of Mid EC control

Table C 3-2. Chemistry of sample waters examined in a *H. transpacificus* (Delta Smelt) 7-day test initiated on 4/24/08, evaluating the toxicity of Sacramento River and Delta water samples collected 4/22/08-4/24/08.

Sample	Collection Date and Time	Field Chemistry				
		SC (uS/cm)	EC (uS/cm)	Temp (°C)	DO (mg/L)	pH
Suisun at Rush Ranch	4/23/2008 10:50	4864	3901	15.1	7.0	7.38
Sacramento River at Hood DWR Station	4/22/2008 13:00	170	142	16.8	9.3	7.75
Sacramento River, Deep Water Channel, Light 55	4/23/2008 10:20	347	284	15.9	10.1	8.36
Confluence of Lindsey Slough and Cache Slough	4/23/2008 11:15	359	291	15.5	10.2	8.29
Rough and Ready DWR Station, Stockton	4/22/2008 10:30	579	494	17.7	7.9	7.89
Napa River at Vallejo Seawall (340)	4/24/2008 12:25	22080	17700	15.1	10.2	7.92

Sample	Turbidity (NTU)	Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)[1]
Suisun at Rush Ranch	74.5	0.14	0.001
Sacramento River at Hood DWR Station	18.8	0.4	0.007
Sacramento River, Deep Water Channel, Light 55	27	0.13	0.008
Confluence of Lindsey Slough and Cache Slough	31.3	0.17	0.008
Rough and Ready DWR Station, Stockton	34.2	0.17	0.004
Napa River at Vallejo Seawall (340)	18.5	0.09	0.001

Table C 3-3. Chemistry of sample waters examined in a *Hypomesus* (Delta Smelt) 7-day test initiated 4/24/08 evaluating the toxicity of Sacramento River and Delta water samples collected 4/22/08 - 4/24/08.

Treatment	Temp (°C)			EC (uS/cm)			DO (mg/L)		
	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*
Low EC Control	16.6	0.3	8	197	57	8	9.1	0.4	8
Suisun at Rush Ranch	16.4	0.4	8	3686	74	8	9.6	0.4	8
Sacramento River at Hood DWR Station	16.2	0.3	8	165	43	8	9.7	0.2	8
Sacramento River, Deep Water Channel, Light 55	16.1	0.3	8	289	34	8	9.8	0.3	8
Confluence of Lindsey Slough and Cache Slough	16.1	0.4	8	259	42	8	9.6	0.4	8
Rough and Ready DWR Station, Stockton	16.3	0.4	8	476	32	8	9.6	0.3	8
Mid EC Control	16.2	0.3	8	478	42	8	9.7	0.3	8
High EC Control	16.3	0.3	8	3237	1198	8	9.7	0.3	8
Napa River at Vallejo Seawall (340)	16.2	0.3	8	17404	1340	8	9.3	0.3	8
Low Turbidity Control	16.6	0.3	8	3728	67	8	9.5	0.2	8

Treatment	pH			Turbidity (NTU)			Ammonia Nitrogen (mg/L)		
	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*
Low EC Control	7.69	0.23	8	4.8	1.7	7	0.348	0.2412	4
Suisun at Rush Ranch	7.95	0.20	8	19.5	4.4	7	0.106	0.0321	5
Sacramento River at Hood DWR Station	7.92	0.17	8	2.7	0.8	7	0.292	0.1152	5
Sacramento River, Deep Water Channel, Light 55	8.10	0.09	8	10.4	1.3	7	0.132	0.0239	5
Confluence of Lindsey Slough and Cache Slough	8.08	0.08	8	11.2	1.9	7	0.156	0.0152	5
Rough and Ready DWR Station, Stockton	8.01	0.10	8	5.6	1.3	7	0.156	0.0195	5
Mid EC Control	8.05	0.09	8	5.3	2.1	7	0.073	0.005	4
High EC Control	8.00	0.08	8	5.0	2.4	7	0.080	0.0082	4
Napa River at Vallejo Seawall (340)	7.81	0.07	8	5.4	1.7	7	0.066	0.0182	5
Low Turbidity Control	7.84	0.15	8	2.6	0.9	7	0.043	0.010	4

Treatment	Unionized Ammonia (mg/L)			Hardness (mg/L as $CaCO_3$)	Alkalinity (mg/L as $CaCO_3$)
	Mean	SD	*N*		
Low EC Control	0.004	0.003	4	-	-
Suisun at Rush Ranch	0.002	0.001	5	632	192
Sacramento River at Hood DWR Station	0.006	0.002	5	56	62
Sacramento River, Deep Water Channel, Light 55	0.004	0.001	5	116	114
Confluence of Lindsey Slough and Cache Slough	0.005	0.001	5	96	108
Rough and Ready DWR Station, Stockton	0.004	0.001	5	124	88
Mid EC Control	0.002	0.000	4	-	-
High EC Control	0.002	0.001	4	-	-
Napa River at Vallejo Seawall (340)	0.001	0.000	5	2368	116
Low Turbidity Control	0.001	0.000	4	-	-

Table C 4-1. Results of a *H. transpacificus* (Delta Smelt) 7-day test initiated 5/22/08 evaluating the toxicity of ambient water samples from the Sacramento River and Delta collected on 5/20/08 - 5/22/08.[1]

Treatment	Survival (%)[2]	
	mean	se
Low EC Control	75.0	2.9
Sacramento River at Hood DWR Station[3,6]	46.1	10.6
Confluence of Lindsey Sl. And Cache Sl.[3]	71.3	10.9
Sacramento R. Deep Water Channel, Light 55[3]	77.5	6.3
Hatchery Control	75.0	9.6
Rough and Ready DWR station, Stockton[4]	55.0	6.5
High EC Control	88.8	5.2
Low Turbidity Control (High EC)	69.2	14.7
Suisun Slough at Rush Ranch[5]	85.0	6.5
Napa River at Vallejo Seawall (340)[5]	92.5	4.8

1. Smelt used in this experiment were 57 days old at test initiation.
2. Highlighted areas indicate significant reductions in survival compared to the appropriate EC-specific control. Data were analyzed using both USEPA standard single concentration statistical protocols and ANOVA with Tukey's multiple comparison procedure.
3. These low conductivity samples were compared to the Low EC control.
4. This intermediate conductivity sample was compared to the hatchery rearing water control.
5. These high conductivity samples were compared to the High EC control.
6. This sample was found to have reduced survival when evaluated by USEPA statistical protocols, but not when evaluated by Tukey's multiple comparison procedure.

	MSD	**PMSD†**
Survival (%)	**37.2**	**49.6**

†. Calculated as % of Hatchery Water control

Table C 4-2. Field chemistry of sample waters examined in a *H. transpacificus* (Delta Smelt) 7-day test initiated on 5/22/08, evaluating the toxicity of Sacramento River and Delta water samples collected on 5/20/08 - 5/22/08.

Sample	Collection Date and Time	SC (uS/cm)	EC (uS/cm)	Temp (°C)	DO (mg/L)	pH
Sacramento River at Hood DWR Station	5/20/2008 12:00	180.2	177	24.1	7.9	7.32
Confluence of Lindsey Sl. And Cache Sl.	5/21/2008 11:00	242.8	216	19.5	8.7	7.22
Sacramento R. Deep Water Channel, Light 55	5/21/2008 9:40	280.3	256	20.7	8.8	7.47
Rough and Ready DWR station, Stockton	5/20/2008 10:00	400.6	385	23.1	7.3	7.08
Suisun Slough at Rush Ranch	5/21/2008 8:20	6870	5757	16.9	6.7	7.38
Napa River at Vallejo Seawall (340)	5/22/2008 10:40	27910	23724	17.5	9.4	7.81

Sample	Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)	Turbidity (NTU)
Sacramento River at Hood DWR Station	0.41	0.004	17
Confluence of Lindsey Sl. And Cache Sl.	0.23	0.001	72
Sacramento R. Deep Water Channel, Light 55	0.17	0.002	40
Rough and Ready DWR station, Stockton	0.07	0.000	12
Suisun Slough at Rush Ranch	0.11	0.001	47
Napa River at Vallejo Seawall (340)	0.15	0.002	114

Table C 4-3. Chemistry of sample waters examined in a *H. transpacificus* (Delta Smelt) 7-day test initiated 5/22/08 evaluating the toxicity of Sacramento River and Delta water samples collected on 5/20/08 - 5/22/08.

Treatment	Temp (°C)			EC (uS/cm)			DO (mg/L)		
	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*
Low EC Control	17.1	0.3	8	187	28	8	8.9	0.3	8
Suisun Slough at Rush Ranch	16.8	0.5	8	5733	250	8	8.7	0.7	8
Sacramento River at Hood DWR Station	16.5	0.5	8	163	28	8	9.3	0.5	8
Sacramento R. Deep Water Channel, Light 55	16.7	0.2	8	239	25	8	9.4	0.2	8
Confluence of Lindsey Sl. And Cache Sl.	16.7	0.4	8	227	30	8	9.7	0.2	8
Rough and Ready DWR station, Stockton	16.7	0.4	8	339	41	8	9.3	0.5	8
Hatchery Control	17.0	0.2	8	369	39	8	9.2	0.2	8
High EC Control	16.9	0.4	8	5825	400	8	9.3	0.2	8
Napa River at Vallejo Seawall (340)	16.6	0.5	8	22443	630	8	8.9	0.3	8
Low Turbidity Control	17.1	0.3	8	5707	235	8	9.3	0.2	8

Treatment	pH			Turbidity (NTU)			Ammonia Nitrogen (mg/L)		
	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*
Low EC Control	7.61	0.21	8	7.8	0.9	6	0.37	0.33	4
Suisun Slough at Rush Ranch	7.67	0.39	8	13.0	1.3	6	0.11	0.03	4
Sacramento River at Hood DWR Station	7.79	0.16	8	4.1	0.6	6	0.25	0.09	4
Sacramento R. Deep Water Channel, Light 55	7.98	0.19	8	14.3	0.4	6	0.18	0.02	4
Confluence of Lindsey Sl. And Cache Sl.	8.02	0.17	8	19.5	0.8	6	0.22	0.02	4
Rough and Ready DWR station, Stockton	7.81	0.08	8	5.1	0.7	6	0.13	0.04	4
Hatchery Control	7.75	0.07	8	6.6	1.0	6	0.28	0.24	4
High EC Control	7.79	0.20	8	4.9	0.7	6	0.12	0.11	4
Napa River at Vallejo Seawall (340)	7.76	0.30	8	10.0	1.7	6	0.06	0.05	4
Low Turbidity Control	7.84	0.25	8	3.2	0.5	6	0.12	0.07	4

Treatment	Unionized Ammonia (mg/L)			Hardness (mg/L as $CaCO_3$)	Alkalinity (mg/L as $CaCO_3$)
	Mean	SD	*N*		
Low EC Control	0.004	0.003	4	-	-
Suisun Slough at Rush Ranch	0.002	0.001	4	800	168
Sacramento River at Hood DWR Station	0.005	0.002	4	64	66
Sacramento R. Deep Water Channel, Light 55	0.005	0.001	4	88	92
Confluence of Lindsey Sl. And Cache Sl.	0.007	0.001	4	80	88
Rough and Ready DWR station, Stockton	0.003	0.001	4	88	64
Hatchery Control	0.004	0.004	4	-	-
High EC Control	0.002	0.002	4	-	-
Napa River at Vallejo Seawall (340)	0.001	0.001	4	3120	118
Low Turbidity Control	0.002	0.0014	4	-	-

Table C 5-1. Results of a *H. transpacificus* (Delta Smelt) 7-day test initiated 3/19/09 evaluating the toxicity of ambient water samples collected on 3/17/09, 3/18/09 and 3/19/09. Test animals were 30 days old at test initiation.

Treatment	Survival (%)		EC-specific Statistical Results[1]
	mean	se	
Low EC Control	8.3	5.3	A
Low EC Low Turbidity Control	2.8	2.8	A
Hood[2]	8.7	2.9	A
Light 55[2]	23.6	9.2	A
Cache Lindsey[2]	2.8	2.8	A
Mid EC Control	15.3	6.4	A
Rough and Ready Island[3]	2.8	2.8	A
High EC Control	18.6	7.9	B
High EC Low Turbidity Control	18.1	6.4	B
Suisun[4]	95.0	5.0	A
340[4]	88.8	4.1	A

1. Data were analyzed using separate statistical tests for each EC bracket (low, mid, high). The low and high EC brackets were examined using Tukey's tests, while the intermediate EC bracket was examined using a T-test (all tests were two-tailed, $\alpha = 0.05$). Statistically different groups of treatments are identified by different letters. Due to the poor performance of the controls, USEPA standard statistics were not performed.

2. These low conductivity samples were compared to the Low EC controls.

3. This intermediate conductivity sample was compared to the Mid EC Control.

4. These high conductivity samples were compared to the High EC controls.

	Survival (%) MSD
Low Conductivity	22.8
Mid Conductivity	17.1
High Conductivity	25.3

Table C 5-2. Field chemistry of sample waters examined in a *H. transpacificus* (Delta Smelt) 7-day test initiated on 3/19/09, evaluating the toxicity of Sacramento River and Delta water samples collected 3/17/09-3/19/09.

Sample	Collection Date and Time	Field Chemistry				
		SC (uS/cm)	EC (uS/cm)	Temp (°C)	DO (mg/L)	pH
Suisun Slough at Rush Ranch	3/18/2009 16:50	4106	3642	19.0	9.2	7.42
Sacramento River at Hood DWR Station	3/19/2009 9:30	197	157	14.4	9.9	7.14
Sacramento R. Deep Water Channel, Light 55	3/18/2009 10:10	369	290	13.0	10.0	7.79
Confluence of Lindsey Sl. And Cache Sl.	3/18/2009 11:10	354	281	14.3	10.0	7.38
Rough and Ready DWR station, Stockton	3/17/2009 16:00	740	604	15.7	8.5	7.68
Napa River at Vallejo Seawall (340)	3/17/2009 10:40	11,210	8,680	13.2	10.3	7.02

Table C 5-3. Chemistry of sample waters examined in a *H. transpacificus* (Delta Smelt) 7-day test initiated 3/19/09 evaluating the toxicity of Sacramento River and Delta water samples collected on 3/17/09 - 3/19/09.

Treatment	Temp (°C)			EC (uS/cm)			SC (uS/cm)			DO (mg/L)		
	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*
Low EC Control	16.3	0.9	8	172	9	8	206	12	8	9.1	0.3	8
Suisun	16.1	1.1	8	3234	148	8	3922	156	8	8.9	0.2	8
Hood	16.0	1.0	8	167	10	8	201	13	8	9.4	0.3	8
Light 55	16.0	1.0	8	296	6	8	357	3	8	9.3	0.2	8
Cache Lindsey	16.0	1.0	8	235	6	8	284	3	8	9.3	0.1	8
Rough and Ready Island	16.1	1.1	8	602	19	8	724	6	8	9.3	0.4	8
Mid EC Control	16.3	0.9	8	661	15	8	792	7	8	8.9	0.4	8
High EC Control	16.4	1.0	8	3192	147	8	3824	139	8	9.0	0.4	8
340	16.2	1.1	8	8531	321	8	10224	262	8	9.1	0.4	8
Low EC Low Turbidity Control	16.3	1.0	8	180	28	8	215	31	8	9.1	0.3	8
Low Turbidity Control	16.3	1.0	8	3247	139	8	3886	146	8	9.1	0.4	8

Treatment	pH			Ammonia Nitrogen (mg/L)			Unionized Ammonia (mg/L)			Turbidity (NTU)			Hardness (mg/L as $CaCO_3$)	Alkalinity (mg/L as $CaCO_3$)
	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*		
Low EC Control	7.81	0.13	8	0.52	0.38	4	0.010	0.008	4	5.57	1.49	7	-	-
Suisun	7.93	0.26	8	0.15	0.02	5	0.004	0.002	5	32.66	2.90	7	620	222
Hood	7.99	0.22	8	0.41	0.07	5	0.011	0.005	5	3.36	0.73	7	72	80
Light 55	8.16	0.16	8	0.19	0.02	5	0.008	0.003	5	7.14	0.86	7	124	124
Cache Lindsey	8.10	0.19	8	0.26	0.04	5	0.009	0.004	5	5.10	0.90	7	100	100
Rough and Ready Island	7.98	0.21	8	0.12	0.03	5	0.003	0.002	5	2.60	0.81	7	176	104
Mid EC Control	7.95	0.06	8	0.35	0.19	4	0.008	0.004	4	5.10	1.40	7	-	-
High EC Control	7.89	0.05	8	0.20	0.06	4	0.004	0.001	4	3.58	1.14	7	-	-
340	7.84	0.09	8	0.12	0.03	5	0.002	0.000	5	10.30	3.73	7	1260	88
Low EC Low Turbidity Control	7.92	0.12	8	0.23	0.10	4	0.006	0.005	4	3.24	1.05	7	-	-
Low Turbidity Control	7.87	0.03	8	0.13	0.03	4	0.002	0.000	4	2.13	1.68	7	-	-

Table C 6-1. Results of a *H. transpacificus* (Delta Smelt) 7-day test initiated 4/02/09 evaluating the toxicity of ambient water samples collected on 3/31/09 - 4/02/09. Test organisms were 44 days old at test initiation.

Treatment	96-hr Survival (%)[1]					7-day Survival (%)[1]				
			USEPA Statistics					USEPA Statistics		
	Mean	SE	v. SC-specific control	v. SC-specific low turbidity control	SC-specific Tukeys Result	Mean	SE	v. SC-specific control	v. SC-specific low turbidity control	SC-specific Tukeys Result
Low SC Control	85.0	6.5	-	-	A	70.0	8.2	-	-	A
Low SC Low Turbidity Control	66.8	5.8	S*	-	A B	43.0	6.0	S*	-	A B
Low SC Control + Tannins	31.8	2.8	S***	S**	C	2.5	2.5	S*	S*	C
Sacramento River at Hood DWR Station[2]	51.0	12.0	S*	NS	B C	19.5	6.1	S**	S*	B C
Sacramento R. Deep Water Channel, Light 55[2]	69.3	5.4	NS	NS	A B	40.7	3.2	S*	NS	A B
Confluence of Lindsey Sl. And Cache Sl.[2]	53.6	8.7	S*	NS	A B C	25.0	11.4	S*	NS	B C
Mid EC Control	81.4	3.7	-	-	A	69.5	4.9	-	-	A
Rough and Ready DWR station, Stockton[3]	43.0	6.5	S**	-	B	9.3	3.7	S***	-	B
High SC Control	86.1	5.8	-	-	A	64.5	12.8	-	-	A
High SC Low Turbidity Control	81.6	13.1	NS	-	A	61.6	11.2	NS	-	A
Suisun Slough at Rush Ranch[4]	97.7	2.3	NS	NS	A	95.5	2.6	NS	NS	A
Napa River at Vallejo Seawall (340)[4]	88.6	8.6	NS	NS	A	74.8	9.2	NS	NS	A

1. Data were analyzed using a separate statistical tests for each EC bracket (low, mid, high), and both standard USEPA statistics (one-tailed $\alpha = 0.05$) and ANOVA with Tukeys multiple comparison (two-tailed $\alpha = 0.05$) were performed. The intermediate EC bracket was examined using a T-test instead of Tukey's test. Statistically different groups of treatments are identified by highlighting (USEPA) and by different letters (Tukey).

*: $P < 0.05$

**: $P < 0.01$

***: $P < 0.001$

2. These low conductivity samples were compared to the Low EC controls.

3. This intermediate conductivity sample was compared to the Mid EC Control.

4. These high conductivity samples were compared to the High EC controls.

	Tukey's test MSD	
	96-hr Survival (%)	7-day Survival (%)
Low Conductivity	29.6	27.5
Mid Conductivity	14.6	11.9
High Conductivity	30.5	35.3

Table C 6-2. Field chemistry of sample waters examined in a *H. transpacificus* (Delta Smelt) 7-day test initiated on 4/02/09, evaluating the toxicity of Sacramento River and Delta water samples collected 3/31/09 - 4/02/09.

Sample	Collection Date and Time	Field Chemistry						
		SC (uS/cm)	EC (uS/cm)	Temp (°C)	DO (mg/L)	pH	Ammonia Nitrogen (mg/L)[1]	Unionized Ammonia (mg/L)[1]
Sacramento River at Hood DWR Station	3/31/2009 8:40	177.8	141.2	14.7	9.9	6.91	0.43	0.001
Sacramento R. Deep Water Channel, Light 55	4/2/2009 10:00	283.4	226.2	14.9	10.1	7.02	0.26	0.001
Confluence of Lindsey Sl. And Cache Sl.	4/2/2009 11:15	271.6	220.0	15.5	9.9	6.90	0.28	0.001
Rough and Ready DWR station, Stockton	3/31/2009 13:20	913	754	16.3	10.1	7.94	0.02	0.000
Suisun Slough at Rush Ranch	3/31/2009 12:45	3805	3166	16.6	9.0	7.45	0.27	0.002
Napa River at Vallejo Seawall (340)	4/1/2009 12:15	18760	15233	15.6	9.6	6.94	0.12	0.000

1. Ammonia nitrogen was measured upon sample receipt, and unionized ammonia was calculated using ammonia measured upon receipt and other chemistry measured at sample collection.

Table C 6-3. Chemistry of sample waters examined in a *H. transpacificus* (Delta Smelt) 7-day test initiated 4/02/09 evaluating the toxicity of Sacramento River and Delta water samples collected on 3/31/09 - 4/02/09.

Treatment	Temp (°C)			EC (uS/cm)			SC (uS/cm)			DO (mg/L)		
	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*
Low SC Control	16.2	0.6	8	165	16	8	197	18	8	9.6	0.3	8
Low SC Low Turbidity Control	16.2	0.6	8	198	46	8	238	54	8	9.7	0.2	8
Low SC Control + Tannins	16.3	0.6	8	174	25	8	208	29	8	9.8	0.2	8
Sacramento River at Hood DWR Station	16.1	0.7	8	165	26	8	199	31	8	9.8	0.3	8
Sacramento R. Deep Water Channel, Light 55	16.2	0.7	8	238	22	8	286	26	8	9.8	0.3	8
Confluence of Lindsey Sl. And Cache Sl.	16.1	0.6	8	227	24	8	272	29	8	9.8	0.3	8
Mid SC Control	16.5	0.3	8	789	21	8	941	22	8	9.6	0.3	8
Rough and Ready DWR station, Stockton	16.1	0.7	8	748	20	8	901	20	8	9.9	0.2	8
High SC Control	16.5	0.3	8	3158	73	8	3776	80	8	9.6	0.3	8
High SC Low Turbidity Control	16.6	0.4	8	3229	77	8	3848	72	8	9.6	0.3	8
Suisun Slough at Rush Ranch	16.2	0.5	8	3063	43	8	3683	53	8	9.5	0.5	8
Napa River at Vallejo Seawall (340)	16.1	0.6	8	15134	543	8	18245	520	8	9.4	0.5	8

Treatment	pH			Ammonia Nitrogen (mg/L)			Unionized Ammonia (mg/L)			Turbidity (NTU)			Hardness (mg/L as $CaCO_3$)	Alkalinity (mg/L as $CaCO_3$)
	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*		
Low SC Control	7.82	0.10	8	0.38	0.29	4	0.006	0.005	4	4.47	1.79	8	-	-
Low SC Low Turbidity Control	7.92	0.10	8	0.17	0.13	4	0.003	0.002	4	3.52	1.24	8	-	-
Low SC Control + Tannins	7.88	0.10	8	0.06	0.05	4	0.001	0.001	4	1.94	0.56	8	-	-
Sacramento River at Hood DWR Station	8.01	0.22	8	0.32	0.09	4	0.011	0.004	4	2.72	1.92	8	64	72
Sacramento R. Deep Water Channel, Light 55	8.11	0.15	8	0.27	0.03	4	0.011	0.001	4	5.19	0.99	7	104	92
Confluence of Lindsey Sl. And Cache Sl.	8.10	0.15	8	0.28	0.04	4	0.010	0.002	4	4.57	0.86	7	92	88
Mid EC Control	8.02	0.12	8	0.14	0.07	4	0.004	0.001	4	8.22	4.76	8	-	-
Rough and Ready DWR station, Stockton	8.12	0.11	8	0.09	0.04	4	0.003	0.002	4	3.21	1.91	8	164	104
High SC Control	7.96	0.07	8	0.18	0.11	4	0.004	0.002	4	6.66	4.89	8	-	-
High SC Low Turbidity Control	8.03	0.07	8	0.13	0.05	4	0.003	0.002	4	2.21	1.92	8	-	-
Suisun Slough at Rush Ranch	8.06	0.24	8	0.11	0.05	4	0.004	0.002	4	78.16	108.08	8	496	176
Napa River at Vallejo Seawall (340)	7.83	0.12	8	0.11	0.02	4	0.002	0.000	4	10.68	11.12	8	1996	94

Table C 7-1. Results of a *H. transpacificus* (Delta Smelt) 7-day test initiated 4/16/09 evaluating the toxicity of ambient water samples collected on 4/14/09 - 4/16/09. Test animals were 54 days old at test initiation.

Treatment	96-hr Survival (%)[1]					7-day Survival (%)[1]				
			USEPA Statistics					USEPA Statistics		
	Mean	SE	v. SC-specific control	v. SC-specific low turbidity control	SC-specific Tukeys Result	Mean	SE	v. SC-specific control	v. SC-specific low turbidity control	SC-specific Tukeys Result
Low EC Control	84.7	2.7	-	-	A	58.9	7.2	-	-	A B
Low EC / Low Turbidity Control	46.7	5.4	S***	-	B	27.4	4.0	S**	-	B
Low EC Control + Antibiotics	65.0	12.6	NS	NS	A B	65.0	12.6	NS	NS	A
Sacramento River at Hood DWR Station	67.0	8.1	S*	NS	A B	30.1	6.6	S*	NS	A B
Sacramento R. Deep Water Channel, Light 55	71.4	10.0	NS	NS	A B	55.8	7.9	NS	NS	A B
Confluence of Lindsey Sl. And Cache Sl.	55.3	2.0	S***	NS	A B	46.9	8.5	NS	NS	A B
Mid EC Control	75.6	3.0	-	-	A	67.5	4.6	-	-	A
Rough and Ready DWR station, Stockton	59.8	7.1	S*	-	A	42.2	3.6	S**	-	B
High EC Control	82.5	4.8	-	-	A B	70.0	5.8	-	-	A B
High EC / Low Turbidity Control	83.3	5.6	NS	-	A B	61.9	3.8	NS	-	B
Suisun Slough at Rush Ranch	94.7	3.1	NS	NS	A	92.2	2.6	NS	NS	A
Napa River at Vallejo Seawall (340)	62.2	10.9	S*	NS	B	62.2	10.9	NS	NS	B

1. Data were analyzed using a separate statistical tests for each EC bracket (low, mid, high). Significant reductions in survival compared to EC-specific controls according to USEPA statistics are indicated by shaded cells, groups of treatments found to be significantly different by Tukey's tests are identified by different letters.

2. These low conductivity samples were compared to the Low EC controls.

3. This intermediate conductivity sample was compared to the Mid EC Control.

4. These high conductivity samples were compared to the High EC controls.

	Tukey's Tests Survival (%) MSDs	
	96-hrs	7-days
Low Conductivity	35.0	36.8
Mid Conductivity	18.9	14.3
High Conductivity	24.8	27.5

Table C 7-2. Field chemistry of sample waters examined in a *H. transpacificus* (Delta Smelt) 7-day test initiated 4/16/09 evaluating the toxicity of Sacramento River and Delta water samples collected on 4/14/09 - 4/16/09 .

Sample	Collection Date and Time	Field Chemistry						
		SC (uS/cm)	EC (uS/cm)	Temp (°C)	DO (mg/L)	pH	Ammonia Nitrogen (mg/L)[1]	Unionized Ammonia (mg/L)[1]
Suisun Slough at Rush Ranch	4/15/2009 14:10	4816	4036	16.9	8.8	7.53	0.46	0.004
Sacramento River at Hood DWR Station	4/16/2009 10:15	147.8	116	14.4	10.0	6.85	0.52	0.001
Sacramento R. Deep Water Channel, Light 55	4/15/2009 12:45	408.8	331	15.5	9.5	7.23	0.14	0.001
Confluence of Lindsey Sl. And Cache Sl.	4/15/2009 14:00	674	543	15.3	11.0	7.23	0.16	0.001
Rough and Ready DWR station, Stockton	4/14/2009 13:50	914	766	16.9	8.0	7.72	0.15	0.002
Napa River at Vallejo Seawall (340)	4/14/2009 13:45	19420	15148	14.0	10.2	6.99	0.59	0.001

Table C 7-3. Chemistry of sample waters examined in a *H. transpacificus* (Delta Smelt) 7-day test initiated 4/16/09 evaluating the toxicity of Sacramento River and Delta water samples collected on 4/14/09 - 4/16/09.

Treatment	Temp (°C)			EC (uS/cm)			SC (uS/cm)			DO (mg/L)		
	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*
Low EC Control	17.0	0.2	8	160	31	8	189	36	8	10.1	0.6	8
Low EC / Low Turbidity Control	17.0	0.3	8	165	31	8	193	34	8	9.8	0.6	8
Low EC Control + Antibiotics	17.0	0.3	8	201	50	8	236	58	8	9.7	0.6	8
Sacramento River at Hood DWR Station	16.7	0.5	8	143	36	8	169	42	8	10.2	0.5	8
Sacramento R. Deep Water Channel, Light 55	16.7	0.4	8	332	6	8	387	18	8	10.0	0.6	8
Confluence of Lindsey Sl. And Cache Sl.	16.6	0.4	8	301	6	8	358	7	8	10.2	0.6	8
Mid EC Control	16.8	0.4	8	760	64	8	897	70	8	9.3	0.4	8
Rough and Ready DWR station, Stockton	16.6	0.4	8	766	10	8	909	8	8	10.8	2.3	8
High EC Control	16.7	0.6	8	4101	139	8	4857	159	8	9.8	0.5	8
High EC / Low Turbidity Control	16.8	0.5	8	4212	114	8	4943	149	8	9.7	0.5	8
Suisun Slough at Rush Ranch	16.7	0.4	8	4036	106	8	4785	97	8	9.9	0.9	8
Napa River at Vallejo Seawall (340)	16.8	0.4	8	15918	209	8	18785	222	8	9.6	0.4	8

Treatment	pH			Ammonia Nitrogen (mg/L)			Unionized Ammonia (mg/L)			Turbidity (NTU)			Hardness (mg/L as $CaCO_3$)	Alkalinity (mg/L as $CaCO_3$)
	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*		
Low EC Control	7.89	0.11	8	0.28	0.16	4	0.006	0.003	4	4.51	0.97	7	-	-
Low EC / Low Turbidity Control	7.89	0.17	8	0.22	0.08	4	0.005	0.002	4	3.47	1.42	7	-	-
Low EC Control + Antibiotics	7.87	0.21	8	0.19	0.02	4	0.005	0.003	4	4.94	0.89	7	-	-
Sacramento River at Hood DWR Station	7.98	0.26	8	0.45	0.09	4	0.017	0.006	4	2.19	0.69	7	52	52
Sacramento R. Deep Water Channel, Light 55	8.20	0.12	8	0.14	0.03	4	0.007	0.002	4	5.58	2.23	7	124	108
Confluence of Lindsey Sl. And Cache Sl.	8.23	0.12	8	0.15	0.04	4	0.008	0.003	4	6.84	1.58	6	114	118
Mid EC Control	8.01	0.12	8	0.34	0.17	4	0.009	0.004	4	4.61	1.43	7	-	-
Rough and Ready DWR station, Stockton	8.13	0.17	8	0.16	0.03	4	0.007	0.002	4	3.20	1.43	8	190	112
High EC Control	7.97	0.08	8	0.29	0.13	4	0.007	0.003	4	5.47	1.26	7	-	-
High EC / Low Turbidity Control	7.95	0.10	8	0.12	0.05	4	0.003	0.001	4	1.78	0.96	7	-	-
Suisun Slough at Rush Ranch	8.15	0.33	8	0.16	0.03	4	0.008	0.002	4	31.39	4.74	7	650	248
Napa River at Vallejo Seawall (340)	7.89	0.14	8	0.11	0.02	4	0.002	0.001	4	7.14	2.65	7	2340	100

Table C 8-1. Results of a *H. transpacificus* (Delta Smelt) 7-day test initiated 4/30/09 evaluating the toxicity of Sacramento River and Delta water samples collected on 4/28/09 - 4/30/09. Smelt were 41 days post hatch at test initiation.

Treatment	96-hr Survival (%)[1]		Comparison to EC-Specific Control	Comparison to EC-specific Low Turbidity Control	7-day Survival (%)[1]		Comparison to EC-Specific Control	Comparison to EC-specific Low Turbidity Control
	Mean	SE			Mean	SE		
Low EC Control: No Antibiotics	79.2	4.8	NS	NS	69.4	5.5	NS	NS
Low EC Control	88.2	7.0	-	-	85.9	8.8	-	-
Low EC / Low Turbidity Control	92.5	4.8	NS	-	85.2	3.0	-	-
Sacramento River at Hood DWR Station[2]	79.5	7.8	NS	NS	55.3	4.4	S*	S**
Sacramento R. Deep Water Channel, Light 55[2]	85.0	5.0	NS	NS	80.2	10.1	NS	NS
Confluence of Lindsey Sl. And Cache Sl.[2]	82.5	6.3	NS	NS	67.5	7.5	NS	S*
Mid-EC Control	88.0	4.6	-	-	76.4	4.6	-	-
Rough and Ready DWR station, Stockton[3]	90.7	6.4	NS	-	88.2	7.0	NS	NS
High EC Control	100.0	0.0	-	-	100.0	0.0	-	-
Low Turbidity Control	88.6	4.4	NS	-	86.1	2.5	-	-
Suisun Slough at Rush Ranch[4]	97.5	2.5	NS	NS	93.1	2.3	NS	NS
Napa River at Vallejo Seawall (340)[4]	97.7	2.3	NS	NS	88.2	7.0	NS	NS

1. Highlighted areas indicate significant reductions in survival, weight or biomass compared to the appropriate EC-specific control. Data were analyzed using both USEPA standard single concentration statistical protocols and ANOVA with Tukey's multiple comparison procedure. Tukey's procedure did not detect any significant differences.

*: $P < 0.05$

**: $P < 0.01$

2. These low conductivity samples were compared to the Low EC control.

3. This intermediate conductivity sample was compared to the Mid-EC control.

4. This high conductivity sample was compared to the High EC control.

	Tukey's Tests Survival (%) MSDs	
	96-hrs	7-days
Low Conductivity	23.1	27.0
Mid Conductivity	15.2	16.1
High Conductivity	9.9	14.0

Table C 8-2. Field chemistry of sample waters examined in a *H. transpacificus* (Delta Smelt) 7-day test initiated 4/30/09 evaluating the toxicity of Sacramento River and Delta water samples collected on 4/28/09 - 4/30/09.

Sample	Collection Date and Time	Field Chemistry						
		SC (uS/cm)	EC (uS/cm)	Temp (°C)	DO (mg/L)	pH	Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
Sacramento River at Hood DWR Station	4/28/2009 14:25	119.9	99.3	16.4	8.7	7.08	0.02	0.000
Sacramento R. Deep Water Channel, Light 55	4/30/2009 11:15	236.4	196.7	16.6	9.4	7.25	0.16	0.001
Confluence of Lindsey Sl. And Cache Sl.	4/30/2009 12:40	246.4	206.0	16.8	9.5	6.80	0.20	0.000
Rough and Ready DWR station, Stockton	4/28/2009 15:05	690	604	18.8	7.5	7.74	0.13	0.002
Suisun Slough at Rush Ranch	4/28/2009 10:40	4772	3789	14.7	8.9	7.01	0.14	0.000
Napa River at Vallejo Seawall (340)	4/29/2009 11:15	24360	19196	14.4	9.7	7.49	0.11	0.001

Table C 8-3. Chemistry of sample waters examined in a *H. transpacificus* (Delta Smelt) 7-day test initiated 4/30/09 evaluating the toxicity of Sacramento River and Delta water samples collected on 4/28/09 - 4/30/09.

Treatment	Temp (°C)			EC (uS/cm)			SC (uS/cm)			DO (mg/L)		
	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*
Low EC Control: No Antibiotics	17.1	1.3	8	158	43	8	182	52	8	9.4	0.2	8
Low EC Control	17.0	1.2	8	171	30	8	199	37	8	9.6	0.2	8
Low EC / Low Turbidity Control	17.0	1.2	8	179	36	8	207	44	8	9.6	0.2	8
Sacramento River at Hood DWR Station	16.9	1.2	8	150	30	8	175	36	8	9.8	0.2	8
Sacramento R. Deep Water Channel, Light 55	16.8	1.5	7	244	24	8	291	30	8	9.8	0.3	8
Confluence of Lindsey Sl. And Cache Sl.	16.7	1.4	8	243	22	8	287	30	8	9.9	0.3	8
Mid-EC Control	17.0	1.2	8	716	21	8	845	35	8	9.6	0.3	8
Rough and Ready DWR station, Stockton	16.8	1.2	8	602	19	8	713	31	8	9.5	0.6	8
High EC Control	17.0	1.2	8	3975	74	8	4698	80	8	9.6	0.4	8
Low Turbidity Control	16.9	1.2	8	3774	362	8	4626	54	8	9.7	0.3	8
Suisun Slough at Rush Ranch	16.9	1.2	8	3863	56	8	4598	112	8	9.5	0.4	8
Napa River at Vallejo Seawall (340)	16.8	1.4	8	19420	637	8	23134	572	8	9.1	0.4	8

Treatment	pH			Ammonia Nitrogen (mg/L)			Unionized Ammonia (mg/L)			Turbidity (NTU)			Hardness (mg/L as $CaCO_3$)	Alkalinity (mg/L as $CaCO_3$)
	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*		
Low EC Control: No Antibiotics	7.77	0.12	8	0.1	0.1	4	0.002	0.002	4	6	2	8	-	-
Low EC Control	7.97	0.20	8	0.1	0.1	4	0.002	0.002	4	7	1	8	-	-
Low EC / Low Turbidity Control	7.95	0.14	8	0.1	0.0	4	0.002	0.002	4	7	1	8	-	-
Sacramento River at Hood DWR Station	7.97	0.06	8	0.0	0.0	4	0.001	0.001	4	5	4	8	48	51
Sacramento R. Deep Water Channel, Light 55	8.02	0.04	8	0.1	0.1	4	0.003	0.003	4	16	7	8	84	78
Confluence of Lindsey Sl. And Cache Sl.	8.06	0.07	8	0.1	0.1	4	0.004	0.003	4	16	5	8	64	74
Mid-EC Control	7.93	0.08	8	0.1	0.1	4	0.003	0.002	4	7	2	8	-	-
Rough and Ready DWR station, Stockton	7.95	0.10	8	0.1	0.1	4	0.003	0.002	4	7	3	8	144	93
High EC Control	7.87	0.05	8	0.1	0.1	4	0.002	0.001	4	6	2	8	-	-
Low Turbidity Control	7.83	0.05	8	0.1	0.1	4	0.001	0.001	4	5	3	8	-	-
Suisun Slough at Rush Ranch	7.90	0.17	8	0.1	0.1	4	0.002	0.001	4	29	17	8	640	158
Napa River at Vallejo Seawall (340)	7.71	0.05	8	0.1	0.1	4	0.001	0.001	4	16	17	8	2880	102

Table C 9-1. Results of a *H. transpacificus* (Delta Smelt) 7-day test initiated 5/14/09 evaluating the toxicity of Sacramento River and Delta water samples collected on 5/12/09 - 5/14/09. Smelt were 41 days post hatch at test initiation.

Treatment	96-hour Survival (%)[1]					7-day Survival (%)[1]				
			USEPA Statistics					USEPA Statistics		
	Mean	SE	v. SC-specific control	v. SC-specific low turbidity control	EC-specific Tukey's Test	Mean	SE	v. SC-specific control	v. SC-specific low turbidity control	EC-specific Tukey's Test
Low EC Control	76.4	9.3	-	-	A B	71.4	11.6	-	-	A
Low EC / Low Turbidity Control	68.8	5.0	NS	-	A B	59.7	7.6	NS	-	A
Sacramento River at Hood DWR Station[2]	62.9	6.0	NS	NS	B	52.3	7.8	NS	NS	A
Sacramento R. Deep Water Channel, Light 55[2]	84.7	6.4	NS	NS	A B	85.5	9.8	NS	NS	A
Confluence of Lindsey Sl. And Cache Sl.[2]	94.7	3.1	NS	NS	A	80.1	5.6	NS	NS	A
Mid-EC Control	80.3	4.5	-	-	A	71.9	3.4	-	-	A
Rough and Ready DWR station, Stockton[3]	56.7*	9.1	S*	-	B	28.1***	7.3	S***	-	B
High EC Control @ 4000 uS/cm	86.4	4.7	-	-	A	80.8	3.9	-	-	A
High EC / Low Turbidity Control	85.4	2.6	NS	-	A	55.2*	10.1	S*	-	A
Suisun Slough at Rush Ranch[4]	80.4	12.8	NS	NS	A	85.7	14.3	NS	NS	A
High EC Control @ 17000 uS/cm	72.1	10.0	-	-	A	62.5	13.0	-	-	A
Napa River at Vallejo Seawall (340)[5]	68.9	5.0	NS	-	A	63.9	3.6	NS	-	A

1. Highlighted areas indicate significant reductions in survival, weight or biomass compared to the appropriate EC-specific control. Data were analyzed using both USEPA standard single concentration statistical protocols and ANOVA with Tukey's multiple comparison procedure. Tukey's procedure did not detect any significant differences.

2. These low conductivity samples were compared to the Low EC control.

3. This intermediate conductivity sample was compared to the Mid-EC control.

4. This high conductivity sample was compared to the High EC control @ 4000 uS/cm.

5. This high conductivity sample was compared to the High EC Control @ 17,000 uS/cm.

	96-hr Survival (%) MSD	7-day Survival (%) MSD
Low EC	24.0	33.4
Mid EC	19.8	15.6
High EC @ 4000 uS/cm	26.5	34.4
High EC @ 17000 uS/cm	21.6	26.1

Table C 9-2. Field chemistry of sample waters examined in a *H. transpacificus* (Delta Smelt) 7-day test initiated 5/14/09 evaluating the toxicity of Sacramento River and Delta water samples collected on 5/12/09 - 5/14/09.

Sample	Collection Date and Time	Field Chemistry						
		SC (uS/cm)	EC (uS/cm)	Temp (°C)	DO (mg/L)	pH	Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
Sacramento River at Hood DWR Station	5/12/2009 15:20	116	103	19.3	11.4	6.89	0.21	0.001
Sacramento R. Deep Water Channel, Light 55	5/14/2009 15:20	261	245	21.8	8.8	7.82	0.17	0.005
Confluence of Lindsey Sl. and Cache Sl.	5/14/2009 14:20	207	190	20.7	11.0	7.29	0.16	0.001
Rough and Ready DWR station, Stockton	5/12/2009 14:30	491	455	21.3	6.9	7.43	0.09	0.001
Suisun Slough at Rush Ranch	5/12/2009 11:50	4863	4279	19.0	9.8	6.51	0.32	0.000
Napa River at Vallejo Seawall (340)	5/13/2009 9:15	16330	13881	17.5	9.2	6.91	0.13	0.000

Table C 9-3. Chemistry of sample waters examined in a *H. transpacificus* (Delta Smelt) 7-day test initiated 5/14/09 evaluating the toxicity of Sacramento River and Delta water samples collected on 5/12/09 - 5/14/09.

Treatment	Temp (°C)			EC (uS/cm)			SC (uS/cm)			DO (mg/L)		
	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*
Low EC Control	17.0	0.3	8	161	16	8	189	18	8	9.5	0.2	8
Low EC / Low Turbidity Control	16.9	0.6	8	198	115	8	224	125	8	9.5	0.2	8
Sacramento River at Hood DWR Station	16.7	0.3	8	164	55	8	390	562	8	9.8	0.2	8
Sacramento R. Deep Water Channel, Light 55	16.9	0.5	8	265	14	8	316	18	8	9.7	0.3	8
Confluence of Lindsey Sl. and Cache Sl.	16.6	0.4	8	226	23	8	268	27	8	9.8	0.2	8
Mid-EC Control	17.0	0.3	8	506	28	8	596	32	8	9.4	0.2	8
Rough and Ready DWR station, Stockton	16.7	0.4	8	454	28	8	541	35	8	9.4	0.6	8
High EC Control @ 4000 uS/cm	17.0	0.3	8	4019	68	8	4773	92	8	9.6	0.2	8
High EC / Low Turbidity Control	17.0	0.5	8	4060	65	8	4810	71	8	9.4	0.3	8
Suisun Slough at Rush Ranch	16.8	0.3	8	3995	65	8	4767	80	8	9.4	0.7	8
High EC Control @ 17000 uS/cm	17.1	0.5	8	14473	204	8	17058	243	8	9.1	0.4	8
Napa River at Vallejo Seawall (340)	16.8	0.3	8	13404	146	8	15951	155	8	9.4	0.4	8

Treatment	pH			Ammonia Nitrogen (mg/L)			Unionized Ammonia (mg/L)			Turbidity (NTU)			Hardness (mg/L as $CaCO_3$)	Alkalinity (mg/L as $CaCO_3$)
	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*		
Low EC Control	7.60	0.25	8	0.2	0.1	4	0.002	0.001	4	6	1	8	-	-
Low EC / Low Turbidity Control	7.80	0.17	8	0.2	0.1	4	0.004	0.005	4	5	2	8	-	-
Sacramento River at Hood DWR Station	7.75	0.23	8	0.2	0.0	4	0.004	0.001	4	12	14	8	44	50
Sacramento R. Deep Water Channel, Light 55	7.98	0.07	8	0.1	0.0	4	0.004	0.002	4	41	25	8	76	72
Confluence of Lindsey Sl. and Cache Sl.	7.98	0.20	8	0.1	0.0	4	0.003	0.002	4	35	39	8	76	74
Mid-EC Control	7.89	0.15	8	0.3	0.1	4	0.006	0.005	4	6	2	8	-	-
Rough and Ready DWR station, Stockton	7.79	0.13	8	0.2	0.0	4	0.004	0.001	4	5	2	8	112	70
High EC Control @ 4000 uS/cm	7.93	0.12	8	0.1	0.1	4	0.003	0.002	4	5	3	8	-	-
High EC / Low Turbidity Control	7.97	0.15	8	0.1	0.0	4	0.003	0.002	4	3	2	8	-	-
Suisun Slough at Rush Ranch	7.80	0.27	8	0.2	0.1	4	0.003	0.002	4	21	17	8	384	198
High EC Control @ 17000 uS/cm	7.90	0.17	8	0.1	0.1	4	0.002	0.002	4	7	2	8	-	-
Napa River at Vallejo Seawall (340)	7.67	0.06	8	0.1	0.0	4	0.001	0.001	4	19	24	8	1920	94

Table C 10-1. Results of a *H. transpacificus* (Delta Smelt) 7-day test initiated 5/28/09 evaluating the toxicity of ambient water samples collected on 5/26/09- 5/28/09. Test animals were 55 days old at test initiation.

Treatment	96-hr Survival (%)		7-day Survival (%)	
	mean	se	mean	se
Low EC Control	79.2	10.7	76.4	10.2
Low EC / Low Turbidity Control	87.5	4.8	75.0	2.9
Sacramento River at Hood DWR Station[2]	89.7	7.1	71.1	4.7
Sacramento R. Deep Water Channel, Light 55[2]	91.9	5.3	86.9	5.1
Confluence of Lindsey Sl. And Cache Sl.[2]	91.3	3.0	81.3	4.4
Mid-EC Control	70.8	8.3	62.8	10.3
Rough and Ready DWR station, Stockton[3]	86.1	8.3	72.8	5.8
High EC Control @ 4000 uS/cm	92.5	2.5	82.5	4.8
High EC / Low Turbidity Control	92.5	4.8	71.4	10.0
Suisun Slough at Rush Ranch[4]	89.2	4.5	86.4	5.4
High EC Control @ 17000 uS/cm	70.8	17.2	68.1	15.8
Napa River at Vallejo Seawall (340)[5]	67.5	4.8	62.5	2.5

1. Highlighted areas indicate significant reductions in survival, weight or biomass compared to the appropriate EC-specific control. Data were analyzed using both USEPA standard single concentration statistical protocols and ANOVA with Tukey's multiple comparison procedure. Neither statistical procedure detected any significant differences.

2. These low conductivity samples were compared to the Low EC control.

3. This intermediate conductivity sample was compared to the Mid-EC control.

4. This high conductivity sample was compared to the High EC control @ 4000 uS/cm.

5. This high conductivity sample was compared to the High EC Control @ 17,000 uS/cm.

	96-hr Survival (%) MSD	7-day Survival (%) MSD
Low EC	29.3	26.2
Mid EC	28.8	28.8
High EC @ 4000 uS/cm	16.1	28.1
High EC @ 17000 uS/cm	54.7	49.0

Table C 10-2. Field chemistry of sample waters examined in a *H. transpacificus* (Delta Smelt) 7-day test initiated on 5/28/09 evaluating the toxicity of Sacramento River and Delta water samples collected 5/26/09 - 5/28/09.

Sample	Collection Date and Time	Field Chemistry				
		SC (uS/cm)	EC (uS/cm)	Temp (°C)	DO (mg/L)	pH
Sacramento River at Hood DWR Station	5/26/2009 13:25	152	139	20.7	8.3	6.55
Sacramento R. Deep Water Channel, Light 55	5/28/2009 10:40	215	198	20.9	8.4	7.39
Confluence of Lindsey Sl. and Cache Sl.	5/28/2009 12:10	118	109	21.1	8.4	7.52
Rough and Ready DWR station, Stockton	5/27/2009 15:30	435	442	25.8	6.0	7.08
Suisun Slough at Rush Ranch	5/26/2009 10:20	4755	4137	18.5	7.1	6.82
Napa River at Vallejo Seawall (340)	5/27/2009 11:25	22870	19943	18.6	8.7	6.58

Table C 10-3. Chemistry of sample waters examined in a *H. transpacificus* (Delta Smelt) 7-day test initiated 5/28/09 evaluating the toxicity of Sacramento River and Delta water samples collected on 5/26/09 - 5/28/09.

Treatment	Temp (°C)			EC (uS/cm)			SC (uS/cm)			DO (mg/L)		
	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*
Low EC Control	16.6	0.3	8	144	8	8	171	11	8	9.4	0.4	8
Low EC / Low Turbidity Control	16.4	0.6	8	197	21	8	233	25	8	9.6	0.3	8
Sacramento River at Hood DWR Station	16.4	0.6	8	151	6	8	180	8	8	9.7	0.5	8
Sacramento R. Deep Water Channel, Light 55	16.6	0.5	8	196	11	8	234	15	8	9.7	0.5	8
Confluence of Lindsey Sl. And Cache Sl.	16.5	0.5	8	173	7	8	207	10	8	9.6	0.5	8
Mid-EC Control	16.5	0.3	8	430	24	8	511	27	8	9.7	0.5	8
Rough and Ready DWR station, Stockton	16.4	0.3	8	384	4	8	459	9	8	9.3	1.0	8
High EC Control @ 4000 uS/cm	16.4	0.4	8	4061	115	8	4847	102	8	9.4	0.4	8
High EC / Low Turbidity Control	16.5	0.5	8	3976	122	8	4740	95	8	9.2	0.2	8
Suisun Slough at Rush Ranch	16.5	0.6	8	3843	83	8	4578	75	8	9.3	0.9	8
High EC Control @ 17000 uS/cm	16.4	0.4	8	17080	2945	8	20368	3576	8	9.0	0.4	8
Napa River at Vallejo Seawall (340)	16.2	0.4	8	18059	744	8	21615	723	8	9.1	0.5	8

Treatment	pH			Ammonia Nitrogen (mg/L)			Unionized Ammonia (mg/L)			Turbidity (NTU)			Hardness (mg/L as $CaCO_3$)	Alkalinity (mg/L as $CaCO_3$)
	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*		
Low EC Control	7.85	0.45	8	0.15	0.10	4	0.005	0.005	4	11.95	8.11	8	-	-
Low EC / Low Turbidity Control	7.97	0.60	8	0.13	0.11	4	0.004	0.006	4	7.62	1.71	8	-	-
Sacramento River at Hood DWR Station	7.90	0.33	8	0.38	0.08	4	0.013	0.010	4	4.93	1.54	8	52	54
Sacramento R. Deep Water Channel, Light 55	7.97	0.24	8	0.19	0.05	4	0.007	0.004	4	18.22	5.82	8	64	66
Confluence of Lindsey Sl. And Cache Sl.	7.93	0.22	8	0.21	0.05	4	0.007	0.004	4	20.07	7.32	8	64	62
Mid-EC Control	8.04	0.21	8	0.16	0.10	4	0.005	0.003	4	10.52	8.81	8	-	-
Rough and Ready DWR station, Stockton	7.87	0.28	8	0.13	0.06	4	0.004	0.003	4	8.48	1.65	8	96	68
High EC Control @ 4000 uS/cm	7.90	0.20	8	0.13	0.06	4	0.002	0.001	4	10.43	8.04	8	-	-
High EC / Low Turbidity Control	7.75	0.40	8	0.09	0.06	4	0.001	0.001	4	3.87	2.47	8	-	-
Suisun Slough at Rush Ranch	7.68	0.31	8	0.13	0.05	4	0.002	0.001	4	20.09	2.66	8	520	142
High EC Control @ 17000 uS/cm	7.95	0.11	8	0.05	0.04	4	0.001	0.001	4	10.5	8.1	8	-	-
Napa River at Vallejo Seawall (340)	7.71	0.12	8	0.07	0.04	4	0.001	0.001	4	10.89	9.30	8	2640	100

Appendix D

Summary Tables:
In-situ Toxicity Tests

Table D1. 96-hour survival of animals examined in flow-through tests initiated at the DWR Station on the Sacramento River at Hood.

Date	Treatment	*H. transpacificus*		*P. promelas*		*H. azteca*	
		Survival (%)		Survival (%)		Survival (%)	
		Mean	SE	Mean	SE	Mean	SE
3/19/2009	Control	-	-	100	0.0	100	0.0
	Ambient	-	-	85	9.6	95	5.0
4/2/2009	Control	75	4.8	85	9.6	95	5.0
	Ambient	84	10.3	90	5.8	85	9.6
4/16/2009	Control	59	5.0	95	5.0	95	5.0
	Ambient	74	10.5	90	5.8	95	5.0
4/30/2009	Control	47	10.9	95	5.0	100	0.0
	Ambient	43	6.5	100	0.0	100	0.0
5/14/2009	Control	56	18.8	95	5.0	100	0.0
	Ambient	69	12.0	100	0.0	100	0.0
5/28/2009	Control	13	8.0	85	5.0	95	5.0
	Ambient	34	7.9	95	5.0	100	0.0

Table D2. 7-day survival of animals examined in flow-through tests at the DWR Station on the Sacramento River at Hood.

Date	Treatment	*H. transpacificus*		*P. promelas*		*H. azteca*	
		Survival (%)		Survival (%)		Survival (%)	
		Mean	SE	Mean	SE	Mean	SE
3/19/2009	Control	21	5.5	85	15.0	95	5.0
	Ambient	46	8.4	75	15.0	95	5.0
4/2/2009	Control	62	8.8	30	5.8	80	8.2
	Ambient	77	7.0	85	5.0	80	8.2
4/16/2009	Control	29	5.1	95	5.0	90	5.8
	Ambient	64	13.8	85	9.6	85	9.6
4/30/2009	Control	43	13.3	95	5.0	100	0.0
	Ambient	40	6.9	95	5.0	100	0.0
5/14/2009	Control	44	15.7	95	5.0	100	0.0
	Ambient	50	10.2	100	0.0	100	0.0
5/28/2009	Control	4	4.2	85	5.0	95	5.0
	Ambient	27	8.4	85	15.0	90	5.8

Table D3. EC and SC chemistry of water from the Sacramento River in exposure chambers during the 7-day in-situ tests at the Hood DWR Station.

Treatment	EC (uS/cm)					SC (uS/cm)				
	Mean	SD	Min	Max	*N*	Mean	SD	Min	Max	*N*
Control Exposure 3/19/2009	153	8	143	162	7	189	8	172	200	14
Ambient Exposure 3/19/2009	140	7	130	153	8	173	7	161	188	13
Control Exposure 4/2/2009	152	13	141	179	7	183	19	159	230	15
Ambient Exposure 4/2/2009	133	11	115	146	7	160	11	140	177	15
Control Exposure 4/16/2009	135	6	126	142	7	153	7	145	167	16
Ambient Exposure 4/16/2009	124	6	117	133	8	148	15	133	194	15
Control Exposure 4/30/2009	111	7	104	123	8	130	7	122	142	14
Ambient Exposure 4/30/2009	104	11	94	119	7	127	9	114	140	15
Control Exposure 5/14/2009	120	7	109	131	8	129	6	118	139	16
Ambient Exposure 5/14/2009	117	12	102	140	8	129	10	114	152	15
Control Exposure 5/28/2009	133	18	111	160	7	148	29	83	211	16
Ambient Exposure 5/28/2009	131	14	114	155	7	137	21	89	170	14

Table D4. Temperature and DO chemistry of water from the Sacramento River in exposure chambers during 7-day in-situ tests at the Hood DWR Station

Treatment	Temperature (ºC)					DO (mg/L)				
	Mean	SD	Min	Max	*N*	Mean	SD	Min	Max	*N*
Control Exposure 3/19/2009	15.4	0.4	15.0	15.9	7	9.3	0.5	8.2	9.8	9
Ambient Exposure 3/19/2009	14.8	0.6	14.0	15.5	7	9.1	0.5	8.0	9.4	8
Control Exposure 4/2/2009	15.9	0.4	15.0	16.0	8	9.2	0.3	8.8	9.6	7
Ambient Exposure 4/2/2009	15.3	0.5	15.0	16.0	8	9.0	0.3	8.4	9.5	7
Control Exposure 4/16/2009	18.3	1.8	16.0	20.0	8	8.9	0.5	8.3	9.6	8
Ambient Exposure 4/16/2009	17.4	1.6	15.0	19.0	8	8.6	0.5	7.8	9.0	7
Control Exposure 4/30/2009	16.3	0.7	15.0	17.0	8	9.4	0.3	8.9	9.8	8
Ambient Exposure 4/30/2009	15.5	0.5	15.0	16.0	8	9.3	0.4	8.5	9.6	7
Control Exposure 5/14/2009	20.9	1.0	19.0	22.0	8	8.4	0.1	8.3	8.6	8
Ambient Exposure 5/14/2009	20.1	0.8	19.0	21.0	8	8.2	0.1	8.1	8.5	7
Control Exposure 5/28/2009	21.8	0.5	21.0	22.0	8	8.2	0.2	8.0	8.7	8
Ambient Exposure 5/28/2009	21.0	0.5	20.0	22.0	8	7.7	0.2	7.4	8.1	7

Table D5. pH and turbidity chemistry of water from the Sacramento River in exposure chambers during 7-day in-situ tests at the Hood DWR Station

Treatment	pH					Turbidity (NTU)				
	Mean	SD	Min	Max	*N*	Mean	SD	Min	Max	*N*
Control Exposure 3/19/2009	7.35	0.36	6.50	7.76	9	2.26	0.91	0.59	3.24	7
Ambient Exposure 3/19/2009	7.52	0.17	7.17	7.73	8	24.99	9.01	15.90	40.20	7
Control Exposure 4/2/2009	7.35	0.11	7.22	7.48	7	2.05	0.66	1.22	3.28	7
Ambient Exposure 4/2/2009	7.40	0.09	7.26	7.49	7	17.80	8.86	6.55	30.20	7
Control Exposure 4/16/2009	7.57	0.23	7.23	7.96	8	3.72	1.86	1.16	6.24	8
Ambient Exposure 4/16/2009	7.52	0.09	7.34	7.61	7	18.39	8.13	9.25	33.80	7
Control Exposure 4/30/2009	7.56	0.11	7.36	7.67	8	5.49	1.33	3.54	6.97	7
Ambient Exposure 4/30/2009	7.56	0.16	7.37	7.81	7	38.53	19.29	10.90	63.20	6
Control Exposure 5/14/2009	7.36	0.14	7.08	7.49	8	4.44	1.07	2.51	5.71	8
Ambient Exposure 5/14/2009	7.32	0.10	7.16	7.46	7	23.31	8.50	12.50	34.30	7
Control Exposure 5/28/2009	7.43	0.10	7.30	7.57	8	2.67	1.08	1.19	4.03	8
Ambient Exposure 5/28/2009	7.37	0.09	7.25	7.50	7	19.20	5.79	13.70	28.30	7

Table D6. Ammonia-nitrogen and unionized ammonia chemistry of water from the Sacramento River in exposure chambers during 7-day in-situ tests at the Hood DWR Station

Treatment	Ammonia Nitrogen (mg/L)					Unionized Ammonia (mg/L)				
	Mean	SD	Min	Max	*N*	Mean	SD	Min	Max	*N*
Control Exposure 3/19/2009	0.21	0.17	0.02	0.49	8	0.002	0.001	0.000	0.003	8
Ambient Exposure 3/19/2009	0.42	0.12	0.28	0.59	7	0.004	0.001	0.002	0.006	7
Control Exposure 4/2/2009	0.15	0.10	0.00	0.26	7	0001	0.001	0.000	0.002	7
Ambient Exposure 4/2/2009	0.42	0.17	0.13	0.64	7	0.003	0.001	0.001	0.005	7
Control Exposure 4/16/2009	0.22	0.17	0.03	0.51	8	0.003	0.002	0.000	0.005	8
Ambient Exposure 4/16/2009	0.37	0.14	0.24	0.66	7	0.004	0.001	0.003	0.005	7
Control Exposure 4/30/2009	0.14	0.11	0.04	0.38	8	0.001	0.001	0.000	0.003	8
Ambient Exposure 4/30/2009	0.22	0.15	0.02	0.45	7	0.002	0.001	0.000	0.004	7
Control Exposure 5/14/2009	0.24	0.16	0.08	0.46	8	0.002	0.002	0.001	0.005	8
Ambient Exposure 5/14/2009	0.29	0.05	0.23	0.35	7	0.002	0.001	0.002	0.004	7
Control Exposure 5/28/2009	0.25	0.18	0.00	0.50	8	0.003	0.002	0.000	0.006	8
Ambient Exposure 5/28/2009	0.32	0.14	0.18	0.53	7	0.003	0.002	0.001	0.006	7

Table D7. Hardness and alkalinity chemistry of water from the Sacramento River in exposure chambers during 7-day in-situ tests at the Hood DWR Station.

Treatment	Hardness (mg/L as $CaCO_3$)					Alkalinity (mg/L as $CaCO_3$)				
	Mean	SD	Min	Max	*N*	Mean	SD	Min	Max	*N*
Control Exposure 3/19/2009	41	3	36	44	7	34	9	18	45	7
Ambient Exposure 3/19/2009	69	3	64	72	7	71	3	68	76	7
Control Exposure 4/2/2009	37	5	32	44	7	32	4	26	36	7
Ambient Exposure 4/2/2009	56	6	48	64	7	62	5	56	68	7
Control Exposure 4/16/2009	34	5	24	40	8	29	5	22	36	8
Ambient Exposure 4/16/2009	52	3	48	56	7	53	8	34	58	7
Control Exposure 4/30/2009	25	4	20	32	7	21	5	12	26	7
Ambient Exposure 4/30/2009	49	3	44	52	6	53	4	48	58	6
Control Exposure 5/14/2009	28	10	12	44	8	23	7	12	32	8
Ambient Exposure 5/14/2009	45	4	40	48	7	47	3	44	52	7
Control Exposure 5/28/2009	32	3	28	36	8	27	3	24	32	8
Ambient Exposure 5/28/2009	50	6	44	60	7	53	4	48	60	7

Table D8. 96-hour survival of animals examined in flow-through tests initiated at the Rough and Ready DWR Station, Stockton, CA.

Date	Treatment	*H. transpacificus*		*P. promelas*		*H. azteca*	
		Survival (%)		Survival (%)		Survival (%)	
		Mean	SE	Mean	SE	Mean	SE
3/19/2009	Control	-	-	94	6.3	95	5.0
	Ambient	-	-	80	0.0	100	0.0
4/2/2009	Control	61	8.9	60	8.2	95	5.0
	Ambient	75	6.8	90	10.0	100	0.0
4/16/2009	Control	63	9.7	65	12.6	100	0.0
	Ambient	71	9.8	45	12.6	100	0.0
4/30/2009	Control	79	12.5	75	9.6	95	5.0
	Ambient	61	16.5	70	12.9	100	0.0
5/14/2009	Control	15	9.6	95	5.0	100	0.0
	Ambient	15	8.6	100	0.0	100	0.0
5/28/2009	Control	-	-	100	0.0	100	0.0
	Ambient	-	-	100	0.0	100	0.0

Table D9. 7-day survival of animals examined in flow-through tests initiated at the Rough and Ready DWR Station, Stockton, CA.

Date	Treatment	*H. transpacificus*		*P. promelas*		*H. azteca*	
		Survival (%)		Survival (%)		Survival (%)	
		Mean	SE	Mean	SE	Mean	SE
3/19/2009	Control	22	10.4	71	16.1	95	5.0
	Ambient	35	9.3	65	9.6	100	0.0
4/2/2009	Control	41	7.9	45	5.0	95	5.0
	Ambient	61	9.4	90	10.0	100	0.0
4/16/2009	Control	59	12.4	65	12.6	100	0.0
	Ambient	66	12.5	40	8.2	100	0.0
4/30/2009	Control	68	15.8	70	12.9	90	10.0
	Ambient	47	14.1	70	12.9	85	9.6
5/14/2009	Control	0	0.0	95	5.0	95	5.0
	Ambient	15	8.6	100	0.0	100	0.0
5/28/2009	Control	-	-	100	0.0	100	0.0
	Ambient	-	-	100	0.0	95	5.0

Table D10. EC and SC chemistry of water in exposure chambers during the 7-day in-situ tests at the Rough and Ready DWR Station in Stockton, CA.

Treatment	EC (uS/cm)					SC (uS/cm)				
	Mean	SD	Min	Max	*N*	Mean	SD	Min	Max	*N*
Control Exposure 3/19/2009	688	98	584	826	8	803	99	720	970	14
Ambient Exposure 3/19/2009	698	72	615	793	8	836	88	730	948	14
Control Exposure 4/2/2009	787	14	765	812	8	915	13	895	944	15
Ambient Exposure 4/2/2009	783	31	740	851	8	915	29	879	1005	15
Control Exposure 4/16/2009	834	37	795	898	7	899	35	849	958	16
Ambient Exposure 4/16/2009	767	19	729	791	7	852	47	790	925	14
Control Exposure 4/30/2009	591	108	482	832	8	569	199	131	947	16
Ambient Exposure 4/30/2009	490	59	429	589	8	546	65	472	668	15
Control Exposure 5/14/2009	479	93	388	696	8	495	99	410	744	16
Ambient Exposure 5/14/2009	423	29	385	461	8	439	29	405	498	15
Control Exposure 5/28/2009	437	25	407	492	8	440	47	290	508	16
Ambient Exposure 5/28/2009	420	5	415	430	8	416	55	275	446	15

Table D11. Temperature and DO chemistry of water in exposure chambers during the 7-day in-situ tests at the Rough and Ready DWR Station in Stockton, CA.

Treatment	Temperature (ºC)					DO (mg/L)				
	Mean	SD	Min	Max	*N*	Mean	SD	Min	Max	*N*
Control Exposure 3/19/2009	16.8	0.9	15.1	17.8	8	9.6	0.4	8.9	10.3	8
Ambient Exposure 3/19/2009	16.2	0.8	15.4	17.8	8	9.0	0.8	7.2	9.6	8
Control Exposure 4/2/2009	17.6	0.5	16.5	18.1	8	9.3	0.4	8.8	9.9	8
Ambient Exposure 4/2/2009	17.0	0.5	16.3	17.5	8	9.4	0.4	8.7	9.9	8
Control Exposure 4/16/2009	19.7	1.9	16.8	22.3	8	8.8	0.5	8.1	9.5	8
Ambient Exposure 4/16/2009	19.0	1.8	16.8	21.8	8	8.7	0.5	7.7	9.1	7
Control Exposure 4/30/2009	19.3	0.5	18.7	20.1	8	8.7	0.2	8.4	8.9	8
Ambient Exposure 4/30/2009	18.7	0.4	18.2	19.5	8	7.2	0.2	7.0	7.5	8
Control Exposure 5/14/2009	23.2	1.0	21.5	24.3	8	8.1	0.3	7.8	8.6	8
Ambient Exposure 5/14/2009	22.7	0.9	21.5	23.9	8	7.1	0.3	6.5	7.4	8
Control Exposure 5/28/2009	24.4	0.2	24.0	24.7	8	8.0	0.2	7.8	8.4	8
Ambient Exposure 5/28/2009	24.0	0.4	23.2	24.4	8	6.1	0.3	5.6	6.5	8

Table D12. pH and turbidity chemistry of water in exposure chambers during 7-day in-situ tests at the Rough and Ready DWR Station in Stockton, CA.

Treatment	pH					Turbidity (NTU)				
	Mean	SD	Min	Max	*N*	Mean	SD	Min	Max	*N*
Control Exposure 3/19/2009	7.83	0.12	7.66	7.98	11	4.42	1.70	2.55	8.26	8
Ambient Exposure 3/19/2009	7.69	0.10	7.56	7.85	11	8.67	2.04	6.67	12.50	8
Control Exposure 4/2/2009	7.56	0.19	7.34	7.84	7	2.87	1.04	1.82	4.79	7
Ambient Exposure 4/2/2009	7385	0.08	7.67	7.91	7	12.29	2.30	8.49	15.90	7
Control Exposure 4/16/2009	7.71	0.08	7.60	7.85	8	2.58	0.87	1.60	4.01	8
Ambient Exposure 4/16/2009	7.94	0.11	7.81	8.14	7	9.72	2.42	7.04	13.80	7
Control Exposure 4/30/2009	7.68	0.22	7.50	8.19	8	4.71	1.29	3.61	7.72	8
Ambient Exposure 4/30/2009	7.55	0.11	7.41	7.66	7	13.87	4.44	11.50	23.90	7
Control Exposure 5/14/2009	7.61	0.07	7.54	7.72	8	2.80	1.08	1.43	4.53	8
Ambient Exposure 5/14/2009	7.48	0.04	7.43	7.53	7	10.83	2.35	8.57	14.60	7
Control Exposure 5/28/2009	7.83	0.15	7.61	8.05	8	6.51	4.35	2.46	16.46	8
Ambient Exposure 5/28/2009	7.37	0.06	7.29	7.46	7	17.29	7.65	9.33	33.23	7

Table D13. Ammonia-nitrogen and unionized ammonia chemistry of water in exposure chambers during 7-day in-situ tests at the Rough and Ready Station in Stockton, CA.

Treatment	Ammonia Nitrogen (mg/L)					Unionized Ammonia (mg/L)				
	Mean	SD	Min	Max	*N*	Mean	SD	Min	Max	*N*
Control Exposure 3/19/2009	0.15	0.12	0.03	0.34	8	0.003	0.003	0.000	0.009	8
Ambient Exposure 3/19/2009	0.07	0.02	0.06	0.10	8	0.001	0.00	0.001	0.001	8
Control Exposure 4/2/2009	0.27	0.18	0.04	0.52	7	0.002	0.001	0.001	0.003	7
Ambient Exposure 4/2/2009	0.08	0.04	0.04	0.17	7	0.001	0.000	0.001	0.002	7
Control Exposure 4/16/2009	0.30	0.15	0.11	0.50	8	0.005	0.003	0.001	0.010	8
Ambient Exposure 4/16/2009	0.09	0.02	0.05	0.11	7	0.002	0.001	0.001	0.003	7
Control Exposure 4/30/2009	0.28	0.17	0.09	0.53	8	0.004	0.002	0.001	0.007	8
Ambient Exposure 4/30/2009	0.10	0.04	0.07	0.18	7	0.001	0.001	0.001	0.003	7
Control Exposure 5/14/2009	0.22	0.16	0.00	0.49	8	0.004	0.003	0.000	0.009	8
Ambient Exposure 5/14/2009	0.10	0.11	0.00	0.29	7	0.001	0.001	0.000	0.004	7
Control Exposure 5/28/2009	0.12	0.12	0.00	0.30	8	0.003	0.003	0.000	0.008	8
Ambient Exposure 5/28/2009	0.10	0.06	0.05	0.22	7	0.001	0.001	0.001	0.002	7

Table D14. Hardness and alkalinity chemistry in exposure chambers during 7-day in-situ tests at the Rough and Ready DWR Station in Stockton, CA.

Treatment	Hardness (mg/L as $CaCO_3$)					Alkalinity (mg/L as $CaCO_3$)				
	Mean	SD	Min	Max	*N*	Mean	SD	Min	Max	*N*
Control Exposure 3/19/2009	142	16	120	164	7	77	11	56	86	7
Ambient Exposure 3/19/2009	178	11	160	190	7	105	3	102	110	7
Control Exposure 4/2/2009	134	8	124	148	7	68	3	64	72	7
Ambient Exposure 4/2/2009	189	3	184	192	7	112	3	107	116	7
Control Exposure 4/16/2009	143	10	128	160	8	75	10	54	84	8
Ambient Exposure 4/16/2009	181	10	168	196	7	111	3	106	114	7
Control Exposure 4/30/2009	103	16	84	132	7	60	9	48	77	7
Ambient Exposure 4/30/2009	125	12	112	144	6	82	4	76	88	6
Control Exposure 5/14/2009	83	15	72	116	8	52	5	44	60	8
Ambient Exposure 5/14/2009	97	6	88	104	7	67	4	60	72	7
Control Exposure 5/28/2009	100	5	92	108	8	71	4	66	76	8
Ambient Exposure 5/28/2009	95	14	64	104	7	65	7	52	70	7

Appendix E

Summary Tables:

Ambient Toxicity Test

with *Eurytemora affinis*

Table E 1-1. Results of an *E. affinis* 7-day test initiated 5/1/09 evaluating the toxicity of ambient water samples collected on 4/28/09 and 4/30/09.

Treatment	Measured Initial EC	Survival (%)[1]	
		Mean	SE
L16 Media @ 1 ppt	1574	90	10.0
L16 Media @ 1000 µS/cm	825	50	16.7
L16 Media @ 500 µS/cm	441	30	15.3
L16 Media @ 250 µS/cm	248	20	13.3
L16 Media @ 100 µS/cm	131	0	0.0
Sacramento R. Deep Water Channel, Light 55	276	50	22.4
Sacramento River at tip of Grand Island (711)	136	20	13.3
Upper Cache Slough at mouth of Ulatis Creek	325	100	0.0
Sacramento River at Hood DWR Station	124	20	13.3

1. Highlighted cells indicate statistically significant reductions in survival or reproduction compared to the L16 media @ 1 ppt. Ambient samples showed no significant decreases in survival compared to the most appropriate conductivity control waters. Data were analyzed using USEPA standard statistical protocols.

Table E 1-2. Water chemistry at field conditions of ambient delta water samples collected on 4/28/09 and 4/30/09.

Treatment	SC (uS/cm)	EC (uS/cm)	Temp (°C)	pH	DO (mg/L)	Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)[1]	Turbidity (NTU)
Light 55 043009	236	196	16.6	7.25	9.4	0.16	0.001	31.2
711 043009	120	101	17.1	6.88	9.8	0.04	0.000	10.1
CU 043009	329	275	16.8	6.88	9.9	0.03	0.000	45.9
Hood 042809	120	99	16.4	7.08	8.7	0.02	0.000	12.7

1: This unionized ammonia reading is based on the ammonia nitrogen measured upon sample receipt and upon the water chemistry measured at sample collection.

Table E 1-3. Water chemistry during an *E. affinis* 7-day test initiated on 5/01/09 evaluating the toxicity of ambient delta water samples collected on 4/28/09 and 4/30/09.

Treatment	Day 0 - Initial					Day 1 - Final				
	SC (uS/cm)	EC (uS/cm)	Temp (°C)	DO (mg/L)	pH	SC (uS/cm)	EC (uS/cm)	Temp (°C)	DO (mg/L)	pH
L16 @ 1 ppt	1920	1574	16.0	10.0	7.99	1930	1567	15.6	9.5	7.87
L 16 @ 1000 µS/cm	1004	825	16.1	9.7	7.97	1003	828	16.3	9.2	7.88
L 16 @ 500 µS/cm	538	441	16.0	9.7	7.97	517	427	16.3	9.9	7.90
L 16 @ 250 µS/cm	304	248	15.8	9.7	8.00	282	232	16.1	9.8	7.85
L 16 @ 100 µS/cm	160	131	15.8	9.6	7.98	129	106	16.0	9.8	7.79
Light 55	335	276	16.2	9.6	8.01	271	225	16.4	9.5	8.05
711	164	136	16.5	10.0	7.90	136	114	17.0	9.5	7.91
CU	393	325	16.4	10.0	8.24	329	276	16.9	9.6	8.17
Hood	150	124	16.5	10.6	8.02	142	121	17.6	9.4	8.00

Appendix F

H. transpacificus

Tests to Determine Effect Concentrations for Select Contaminants

Table F1-1. Results of a *H. transpacificus* (Delta Smelt) 96-hr test initiated 5/14/08 evaluating the toxicity of copper.

Treatment	Initial Total Copper Measured Conc. (ppb)	Initial Dissolved Copper Measured Conc. (ppb)	96-hour Survival (%)[1,2]	
			mean	se
Hatchery Tap Water (HTW)	2.76	2.53	71.3	3.4
HTW + 37.5 ppb Copper	40.4	41.4	77.5	7.5
HTW + 75 ppb Copper	78.2	76.2	47.5	4.8
HTW + 150 ppb Copper	156	153	0.0	0.0
HTW + 300 ppb Copper	312	304	2.5	2.5
HTW + 600 ppb Copper	608	599	0.0	0.0

1. Highlighted areas indicate significant reductions in survival compared to the hatchery tap water control. Data were analyzed using USEPA standard multiple concentration statistical protocols.

2. Smelt used in this experiment were 49 days old at test initiation.

Table F1-2. Nominal and measured 96-hour effect concentrations for copper (ppb) in a larval *H. transpacificus* (delta smelt) test initiated on 5/14/08.

Endpoint	NOEC	LOEC	96-h LC10		96-h LC50		PMSD
			Estimate	95% C.I.	Estimate	95% C.I.	
Nominal Copper (ppb)	37.5	75	45	19 - 55	85	76 - 95	23.5
Measured Total Copper (ppb)	40.4	78.2	49.7	39.9 - 82.3	97.3	76.7 - 113.3	23.5
Measured Dissolved Copper (ppb)	41.4	76.2	50.1	40.9 - 80.1	92.1	74.7 - 110.8	23.5

Table F1-3. Results of a *H. transpacificus* (Delta Smelt) 96-hr test initiated 5/14/08 evaluating the toxicity of cyfluthrin and bifenthrin.

Treatment	Initial Measured Conc. (ppb)	48-hour Final Measured Conc. (ppb)	96-hour Survival (%)[1,2]	
			mean	se
Hatchery Tap Water (HTW)	-	-	71.3	3.4
HTW Solvent Control	ND / ND	ND / ND	62.5	11.1
HTW + 0.125 ppb Cyfluthrin	0.126	0.003	82.5	7.5
HTW + 0.25 ppb Cyfluthrin	0.240	0.005	52.5	18.0
HTW + 0.5 ppb Cyfluthrin	0.407	0.008	45.7	10.2
HTW + 1.0 ppb Cyfluthrin	0.890	0.017	0.0	0.0
HTW + 2.0 ppb Cyfluthrin	1.500	0.050	0.0	0.0
HTW + 0.125 ppb Bifenthrin	0.065	0.005	80.2	5.9
HTW + 0.25 ppb Bifenthrin	0.120	0.011	54.3	13.8
HTW + 0.5 ppb Bifenthrin	0.260	0.020	2.5	2.5
HTW + 1.0 ppb Bifenthrin	0.500	0.043	0.0	0.0
HTW + 2.0 ppb Bifenthrin	1.100	0.140	0.0	0.0

1. Highlighted areas indicate significant reductions in survival compared to the solvent control. Data were analyzed using USEPA standard multiple concentration statistical protocols.

2. Smelt used in this experiment were 49 days old at test initiation.

Table F1-4. Nominal and measured 96-hour effect concentrations for cyfluthrin (ppb) in a larval *H. transpacificus* (delta smelt) test initiated on 5/14/08.

Endpoint	NOEC	LOEC	96-h LC10		96-h LC50		PMSD
			Estimate	95% C.I.	Estimate	95% C.I.	
Nominal Cyfluthrin (ppb)	0.5	1.0	0.160	0.129 - 0.466	0.454	0.393 - 0.523	68.9
Measured Cyfluthrin (ppb)	0.407	0.890	0.260	0.067 - 0.357	0.420	0.261 - 0.558	36.7

Table F1-5. Nominal and measured 96-hour effect concentrations for bifenthrin (ppb) in a larval *H. transpacificus* (delta smelt) test initiated on 5/14/08.

Endpoint	NOEC	LOEC	96-h LC10		96-h LC50		PMSD
			Estimate	95% C.I.	Estimate	95% C.I.	
Nominal Bifenthrin (ppb)	0.25	0.5	0.215	0.140 - 0.262	0.305	0.246 - 0.359	52.7
Measured Bifenthrin (ppb)	0.120	0.260	0.095	0.061 - 0.117	0.143	0.116 - 0.169	38.7

Table F1-6. Chemistry of sample waters examined in a *H. transpacificus* (Delta Smelt) 4-day test initiated 5/14/08 evaluating the toxicity of copper, cyfluthrin and bifenthrin.

Treatment	Temp (°C)			EC (uS/cm)			DO (mg/L)		
	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*
Hatchery Tap Water (HTW)	18.1	1.2	4	750	-	1	9.3	0.5	4
HTW Solvent Control	17.1	1.1	4	747	-	1	8.6	1.6	4
HTW + 37.5 ppb Copper	17.8	0.7	4	746	-	1	9.4	0.3	4
HTW + 75 ppb Copper	17.8	0.7	4	748	-	1	9.4	0.2	4
HTW + 150 ppb Copper	18.0	0.6	4	757	-	1	9.4	0.3	4
HTW + 300 ppb Copper	18.0	0.8	4	741	-	1	9.4	0.2	4
HTW + 600 ppb Copper	18.2	1.2	3	748	-	1	9.5	0.2	3
HTW + 0.125 ppb Cyfluthrin	17.0	0.9	4	744	-	1	9.2	0.6	4
HTW + 0.25 ppb Cyfluthrin	17.2	1.1	4	753	-	1	8.4	1.9	4
HTW + 0.5 ppb Cyfluthrin	17.1	1.0	4	749	-	1	8.9	1.1	4
HTW + 1.0 ppb Cyfluthrin	17.1	0.8	4	750	-	1	8.5	1.7	4
HTW + 2.0 ppb Cyfluthrin	17.1	0.6	4	757	-	1	8.5	1.7	4
HTW + 0.125 ppb Bifenthrin	16.9	0.6	4	751	-	1	9.3	0.5	4
HTW + 0.25 ppb Bifenthrin	16.8	0.6	4	752	-	1	8.9	1.2	4
HTW + 0.5 ppb Bifenthrin	17.3	1.1	4	748	-	1	8.6	1.6	4
HTW + 1.0 ppb Bifenthrin	16.9	0.6	4	752	-	1	8.7	1.7	4
HTW + 2.0 ppb Bifenthrin	17.2	0.6	4	752	-	1	8.8	1.5	4

Treatment	pH			Ammonia Nitrogen (mg/L)			Un-ionized Ammonia (mg/L)		
	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*
Hatchery Tap Water (HTW)	7.98	0.23	4	0.06	0.05	4	0.001	0.001	4
HTW Solvent Control	7.84	0.26	4	0.03	0.03	4	0.000	0.000	4
HTW + 37.5 ppb Copper	7.92	0.26	4	0.05	0.05	4	0.001	0.001	4
HTW + 75 ppb Copper	7.90	0.34	4	0.05	0.05	4	0.001	0.001	4
HTW + 150 ppb Copper	7.97	0.18	4	0.04	0.04	4	0.001	0.001	4
HTW + 300 ppb Copper	7.87	0.21	4	0.03	0.04	4	0.001	0.001	4
HTW + 600 ppb Copper	7.82	0.42	3	0.05	0.06	4	0.000	0.000	3
HTW + 0.125 ppb Cyfluthrin	7.94	0.14	4	0.01	0.02	4	0.000	0.000	4
HTW + 0.25 ppb Cyfluthrin	7.83	0.36	4	0.01	0.02	4	0.000	0.000	4
HTW + 0.5 ppb Cyfluthrin	7.88	0.22	4	0.01	0.02	4	0.000	0.000	4
HTW + 1.0 ppb Cyfluthrin	7.80	0.27	4	0.01	0.02	4	0.000	0.000	4
HTW + 2.0 ppb Cyfluthrin	7.83	0.33	4	0.03	0.04	4	0.000	0.000	4
HTW + 0.125 ppb Bifenthrin	7.95	0.11	4	0.01	0.02	4	0.000	0.000	4
HTW + 0.25 ppb Bifenthrin	7.86	0.24	4	0.01	0.02	4	0.000	0.000	4
HTW + 0.5 ppb Bifenthrin	7.85	0.29	4	0.01	0.02	4	0.000	0.000	4
HTW + 1.0 ppb Bifenthrin	7.88	0.33	4	0.01	0.02	4	0.000	0.000	4
HTW + 2.0 ppb Bifenthrin	7.86	0.32	4	0.01	0.02	4	0.000	0.000	4

1. The matrix tested was water from the UC Davis Smelt Hatchery, Tracy, CA (Turbidity: 0.64 NTU, Hardness: 164 mg/L, Alkalinity: 58 mg/L).

Table F2-1. Results of a *H. transpacificus* (Delta Smelt) 7-day test initiated 7/08/09 evaluating the toxicity of ammonia. Test animals were 47 days old at test initiation.

Treatment	Mean Measured Ammonia (mg/L)		96-hr Survival (%)		7-day Survival (%)	
	Total Ammonia Nitrogen	Unionized Ammonia	Mean	SE	Mean	SE
Filtered Hatchery Water @ 900 uS/cm	0.1	0.002	67.5	13.1	15.0	8.7
2.5 ppm Ammonium Chloride	1.9	0.032	75.0	18.9	22.5	7.5
5 ppm Ammonium Chloride	3.7	0.064	80.0	9.1	22.5	4.8
10 ppm Ammonium Chloride	7.1	0.099	61.1	3.2	2.5	2.5
20 ppm Ammonium Chloride	14.4	0.191	27.5	8.5	0.0	0.0
40 ppm Ammonium Chloride	29.0	0.333	0.0	0.0	0.0	0.0
80 ppm Ammonium Chloride	57.8	0.645	0.0	0.0	0.0	0.0

1. The 96-hour endpoint was analyzed using USEPA standard multiple concentration statistical protocols.

Table F2-2. Nominal and measured 96-h effect concentrations of ammonia in a *H. transpacificus* test initiated on 7/08/09.

Analyte	NOEC	LOEC	96-hour LC10		96-hour LC50	
			Estimate	95% C.I.	Estimate	95% C.I.
Nominal NH_4Cl (mg/L)	20	40	7.44	< 2.5 - 13.25	16.45	11.35 - 25.57
Measured Ammonia Nitrogen (mg/L)	14.4	29.0	5.38	< 1.9 - 9.38	11.81	8.09 - 18.47
Measured Unionized Ammonia (mg/L)	0.191	0.333	0.084	< 0.002 - 0.127	0.164	0.119 - 0.239

Table F2-3. Water chemistry during a 7-day *H. transpacificus* toxicity test initiated on 7/8/09 examining the toxicity of esfenvalerate and permethrin.

Treatment	Temp (°C)			EC (uS/cm)			DO (mg/L)		
	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*
Filtered Hatchery Water @ 900 uS/cm	17.3	0.5	8	758	-	1	8.9	0.7	8
2.5 ppm Ammonium Chloride	17.2	0.6	8	777	-	1	8.8	0.9	8
5 ppm Ammonium Chloride	17.3	0.8	7	792	-	1	8.8	0.7	8
10 ppm Ammonium Chloride	17.2	0.4	8	820	-	1	8.8	0.9	8
20 ppm Ammonium Chloride	17.1	0.5	7	882	-	1	9.0	0.7	7
40 ppm Ammonium Chloride	17.0	0.2	4	1017	-	1	9.3	0.6	4
80 ppm Ammonium Chloride	16.9	0.0	2	1264	-	1	9.4	0.1	2

Treatment	pH			Ammonia Nitrogen (mg/L)			Un-ionized Ammonia (mg/L)		
	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*
Filtered Hatchery Water @ 900 uS/cm	7.84	0.18	8	0.09	0.06	8	0.002	0.001	8
2.5 ppm Ammonium Chloride	7.76	0.19	8	1.88	0.08	8	0.032	0.013	8
5 ppm Ammonium Chloride	7.75	0.19	8	3.74	0.13	8	0.064	0.027	8
10 ppm Ammonium Chloride	7.67	0.16	8	7.08	0.72	8	0.099	0.040	8
20 ppm Ammonium Chloride	7.66	0.13	7	14.43	0.49	7	0.191	0.056	7
40 ppm Ammonium Chloride	7.61	0.11	4	28.95	2.22	4	0.333	0.100	4
80 ppm Ammonium Chloride	7.62	0.04	2	57.80	4.81	2	0.645	0.105	2

1. The matrix tested was water from the UC Davis Smelt Hatchery, Tracy, CA (Hardness: 100 mg/L, Alkalinity: 66 mg/L).

Table F-2-4. Results of a larval delta smelt LC50 test initiated 6/24/09 evaluating the toxicity of ammonium-chloride. Test animals were 51 days old at test initiation.

Treatment	Measured Mean Values		96-hr Survival (%)	
	Total Ammonia/um (mg/L)	Un-ionized Ammonia (mg/L)	Mean	SE
Filtered Hatchery Water @ 900 uS/cm	0.1	0.001	67.5	14.9
2.5 ppm Ammonia/um	1.8	0.028	57.5	21.7
5 ppm Ammonia/um	3.2	0.050	60.0	16.8
10 ppm Ammonia/um	7.1	0.091	72.5	2.5
20 ppm Ammonia/um	16.3	0.189	12.5	7.5
40 ppm Ammonia/um	32.3	0.281	0.0	0.0
80 ppm Ammonia/um	66.0	0.420	0.0	0.0
Hatchery Water no antibiotics	0.1	0.001	50.0	10.8
10 ppm Ammonia/um no antibiotics	6.8	0.088	42.5	12.5
20 ppm Ammonia/um no antiobiotics	16.0	0.173	15.0	5.0

Table F-2-5. Effect concentrations for 4 d exposures of 51-d old delta smelt larvae to ammonium-chloride.

Point Estimate	Nominal Total Ammonia/um (mg/L)		Measured Total Ammonia/um (mg/L)		Un-ionized Ammonia (mg/L)	
	Estimate	95% C.I.	Estimate	95% C.I.	Estimate	95% C.I.
96-hr Survival LC10	10.37	< 2.5 - 11.54	7.46	< 1.8 - 8.49	0.096	< 0.001 - 0.111
96-hr Survival LC25	11.93	< 2.5 - 13.63	8.83	< 1.8 - 10.35	0.115	< 0.001 - 0.134
96-hr Survival LC50	15.03	9.18 - 18.84	11.63	6.36 - 15.2	0.147	0.085 - 0.181
96-hr NOEC	10	-	7.14	-	0.091	-
96-hr LOEC	20	-	16.31	-	0.189	-

Table F-2-6. Water quality parameters measured during the 4-day test initiated 6/24/09 with 51-d old delta smelt.

Treatment	Temp (°C)			DO (mg/L)			pH			EC (uS/cm)	SC (uS/cm)
	Mean	SD	N	Mean	SD	N	Mean	SD	N		
Filtered Hatchery Water @ 900 uS/cm	17.8	1.5	5	9.1	0.6	5	7.70	0.12	5	767	924
2.5 ppm Ammonia/um	18.1	1.7	5	9.2	0.7	5	7.70	0.14	5	782	956
5 ppm Ammonia/um	17.8	1.6	5	9.0	0.8	5	7.68	0.16	5	791	944
10 ppm Ammonia/um	17.7	1.2	5	8.8	1.1	5	7.60	0.18	5	832	1006
20 ppm Ammonia/um	17.7	1.2	5	9.0	0.9	5	7.58	0.10	5	898	1075
40 ppm Ammonia/um	17.1	1.0	2	8.5	1.0	2	7.49	0.08	2	1007	1204
80 ppm Ammonia/um	16.7	1.1	2	8.8	0.8	2	7.38	0.06	2	1247	1508
Filtered Hatchery Water no antibiotics	17.6	1.3	5	8.5	0.9	5	7.70	0.25	5	726	880
10 ppm Ammonia/um no antibiotics	17.7	0.9	5	8.6	0.8	5	7.62	0.15	5	816	970
20 ppm Ammonia/um no antibiotics	17.6	1.1	5	8.6	0.8	5	7.55	0.12	5	870	1047

Treatment	Ammonia Nitrogen (mg/L)			Unionized Ammonia (mg/L)			Hardness (mg/L as $CaCO_3$)	Alkalinity (mg/L as $CaCO_3$)	Turbidity (NTU)
	Mean	SD	N	Mean	SD	N			
Filtered Hatchery Water @ 900 uS/cm	0.09	0.06	5	0.001	0.001	5	100	79	0.74
2.5 ppm Ammonia/um	1.81	0.09	5	0.028	0.008	5	-	-	-
5 ppm Ammonia/um	3.18	0.90	5	0.050	0.026	5	-	-	-
10 ppm Ammonia/um	7.14	1.12	5	0.091	0.049	5	-	-	-
20 ppm Ammonia/um	16.31	2.21	5	0.189	0.064	5	-	-	-
40 ppm Ammonia/um	32.26	6.70	2	0.281	0.091	2	-	-	-
80 ppm Ammonia/um	65.96	11.37	2	0.420	0.098	2	-	-	-
Filtered Hatchery Water, no antibiotics	0.09	0.08	5	0.001	0.001	5	-	-	-
10 ppm Ammonia/um, no antibiotics	6.85	0.86	5	0.088	0.034	5	-	-	-
20 ppm Ammonia/um, no antibiotics	16.01	2.32	5	0.173	0.062	5	-	-	-

Table F3-1. Results of a 96-hour acute *H. transpacificus* test initiated on 4/07/10 examining the toxicity of esfenvalerate and permethrin.

Treatment	Measured Pesticide (ppb)	96-hour Survival (%)[1]	
		Mean	SE
Hatchery Tap Water (HTW)	-	95	3
HTW Solvent Control	ND / ND	100	0
HTW + 0.075 ppb Esfenvalerate	0.030	93	5
HTW + 0.150 ppb Esfenvalerate	0.051	93	3
HTW + 0.300 ppb Esfenvalerate	0.135	43	8
HTW + 0.600 ppb Esfenvalerate	0.261	0	0
HTW + 1.200 ppb Esfenvalerate	0.628	0	0
Hatchery Tap Water (HTW)	-	98	3
HTW + 1.25 ppb Permethrin	-	98	3
HTW + 2.5 ppb Permethrin	1.374	100	0
HTW + 5.0 ppb Permethrin	2.557	88	3
HTW + 10.0 ppb Permethrin	4.84	35	10
HTW + 20.0 ppb Permethrin	12.88	3	3
HTW + 40.0 ppb Permethrin	24.94	0	0

1. Highlighted cells indicate significantly reduced survival relative to the solvent control. Data were analyzed using USEPA standard single concentration statistical protocols.

Table F3-2. Isomers of permethrin detected in a 96-hour acute *H. transpacificus* test initiated on 4/07/10.

Treatment	Measured Concentration (ppb)
HTW + 2.5 ppb Permethrin	1.374 (0.520 cis, 0.854 trans)
HTW + 5.0 ppb Permethrin	2.557 (0.987 cis, 1.57 trans)
HTW + 10.0 ppb Permethrin	4.84 (1.89 cis, 2.95 trans)
HTW + 20.0 ppb Permethrin	12.88 (4.87 cis, 8.01 trans)
HTW + 40.0 ppb Permethrin	24.94 (9.54 cis, 15.4 trans)

Table F3-3. Nominal 96-hour effect concentrations of esfenvalerate and permethrin in a *H. transpacificus* test initiated on 4/07/10.

Pesticide	Endpoint	NOEC	LOEC	LC10		LC50		PMSD
				Estimate	95% C.I.	Estimate	95% C.I.	
Esfenfvalerate (ppb)	96-hr Survival	0.150	0.300	0.165	0.144 - 0.180	0.280	0.228 - 0.366	13.8%
Permethrin (ppb)	96-hr Survival	5	10	4.695	3.127 - 5.417	8.295	6.658 - 11.220	12.2%

Table F3-4. Measured 96-hour effect concentrations of esfenvalerate and permethrin in a *H. transpacificus* test initiated on 4/07/10.

Pesticide	Endpoint	NOEC	LOEC	LC10		LC50		PMSD
				Estimate	95% C.I.	Estimate	95% C.I.	
Esfenfvalerate (ppb)	96-hr Survival	0.051	0.135	0.054	0 - 0.058	0.117	0.088 - 0.159	11.3%
Permethrin (ppb)	96-hr Survival	2.557	4.84	2.373	1.783 - 2.825	4.065	3.427 - 5.497	12.5%

Table F3-5. Water chemistry during a 96-hour acute *H. transpacificus* toxicity test initiated on 4/7/10 examining the toxicity of esfenvalerate and permethrin.

Treatment	Temp (°C)			EC (uS/cm)			DO (mg/L)		
	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*
Hatchery Tap Water (HTW)	15.5	0.2	4	749	18	2	9.6	0.3	4
HTW Solvent Control	15.4	0.3	4	751	21	2	9.3	0.4	4
HTW + 0.075 ppb Esfenvalerate	15.5	0.3	4	737	40	2	9.2	0.7	4
HTW + 0.150 ppb Esfenvalerate	15.5	0.3	4	747	23	2	9.4	0.4	4
HTW + 0.300 ppb Esfenvalerate	15.5	0.3	4	739	18	2	9.2	0.5	4
HTW + 0.600 ppb Esfenvalerate	15.4	0.3	4	743	33	2	9.2	0.5	4
HTW + 1.200 ppb Esfenvalerate	15.3	0.3	3	743	31	2	9.5	0.3	3
Hatchery Tap Water (HTW)	15.5	0.4	4	742	28	2	9.6	0.2	4
HTW + 1.25 ppb Permethrin	15.5	0.3	4	748	32	2	9.4	0.3	4
HTW + 2.5 ppb Permethrin	15.4	0.2	4	746	35	2	9.3	0.5	4
HTW + 5.0 ppb Permethrin	15.4	0.3	4	744	33	2	9.4	0.6	4
HTW + 10.0 ppb Permethrin	15.4	0.2	4	741	32	2	9.3	0.5	4
HTW + 20.0 ppb Permethrin	15.5	0.2	4	738	41	2	9.2	0.5	4
HTW + 40.0 ppb Permethrin	15.4	0.3	3	740	38	2	9.6	0.3	3

Treatment	pH			Ammonia Nitrogen (mg/L)			Un-ionized Ammonia (mg/L)		
	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*
Hatchery Tap Water (HTW)	8.06	0.10	4	0.11	0.07	3	0.004	0.003	3
HTW Solvent Control	8.00	0.10	4	0.01	0.01	2	0.000	0.000	2
HTW + 0.075 ppb Esfenvalerate	8.02	0.13	4	0.00	0.00	2	0.000	0.000	2
HTW + 0.150 ppb Esfenvalerate	8.02	0.11	4	0.01	0.01	2	0.000	0.000	2
HTW + 0.300 ppb Esfenvalerate	8.01	0.12	4	0.00	0.00	2	0.000	0.000	2
HTW + 0.600 ppb Esfenvalerate	8.01	0.12	4	0.00	-	1	0.000	-	1
HTW + 1.200 ppb Esfenvalerate	8.08	0.11	3	0.00	-	1	0.000	-	1
Hatchery Tap Water (HTW)	8.08	0.10	4	0.11	0.15	2	0.003	0.005	2
HTW + 1.25 ppb Permethrin	8.02	0.13	4	0.00	0.00	2	0.000	0.000	2
HTW + 2.5 ppb Permethrin	8.08	0.14	4	0.01	0.01	2	0.000	0.000	2
HTW + 5.0 ppb Permethrin	8.03	0.12	4	0.00	0.00	2	0.000	0.000	2
HTW + 10.0 ppb Permethrin	8.02	0.13	4	0.00	0.00	2	0.000	0.000	2
HTW + 20.0 ppb Permethrin	8.02	0.13	4	0.00	-	1	0.000	-	1
HTW + 40.0 ppb Permethrin	8.07	0.12	3	0.00	-	1	0.000	-	1

1. The matrix tested was water from the UC Davis Smelt Hatchery, Tracy, CA (Hardness: 164 mg/L, Alkalinity: 92 mg/L).

Table F4-1. Results of a 96-hour acute *H. transpacificus* test initiated on 5/05/10 examining the toxicity of chlorpyrifos.

Treatment	Measured Chlorpyrifos (ppb)	96-hour Survival (%)[1]	
		Mean	SE
Hatchery Tap Water (HTW)	-	95	3
HTW Solvent Control	ND	88	5
HTW + 6.25 ppb Chlorpyrifos	-	92	5
HTW + 12.5 ppb Chlorpyrifos	8.25	90	4
HTW + 25 ppb Chlorpyrifos	14.8	77	5
HTW + 50 ppb Chlorpyrifos	25.2	47	8
HTW + 100 ppb Chlorpyrifos	66.4	28	5
HTW + 200 ppb Chlorpyrifos	156	8	3
HTW + 400 ppb Chlorpyrifos	-	18	8

1. Highlighted cells indicate significantly reduced survival relative to the solvent control. Data were analyzed using USEPA standard single concentration statistical protocols.

Table F4-2. Nominal and measured 96-hour effect concentrations of chlorpyrifos (ppb) in a *H. transpacificus* test initiated on 5/05/10.

Endpoint	NOEC	LOEC	LC10		LC50		PMSD
			Estimate	95% C.I.	Estimate	95% C.I.	
Nominal Chlorpyrifos (ppb)	25	50	13.8	< 6.25 - 38.8	54.7	34.5 - 86.4	21.3%
Measured Chlorpyrifos (ppb)	14.8	25.2	12.8	7.7 - 17.9	27.7	19.0 - 51.2	18.4%

Table F4-3. Water chemistry during a 96-hour acute *H. transpacificus* toxicity test initiated on 5/05/10 examining the toxicity of chlorpyrifos.

Treatment	Temp (°C)			EC (uS/cm)			DO (mg/L)		
	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*
Hatchery Tap Water (HTW)	16.4	0.2	4	750	-	1	9.0	0.7	3
HTW Solvent Control	16.4	0.2	4	747	-	1	9.3	0.8	4
HTW + 6.25 ppb Chlorpyrifos	16.4	0.2	4	751	-	1	9.3	0.7	4
HTW + 12.5 ppb Chlorpyrifos	16.4	0.4	4	747	-	1	9.3	0.8	4
HTW + 25 ppb Chlorpyrifos	16.3	0.3	4	742	-	1	9.3	0.7	4
HTW + 50 ppb Chlorpyrifos	16.3	0.2	4	741	-	1	9.3	0.8	4
HTW + 100 ppb Chlorpyrifos	16.3	0.2	4	742	-	1	9.3	0.7	4
HTW + 200 ppb Chlorpyrifos	16.4	0.2	4	744	-	1	9.3	0.8	4
HTW + 400 ppb Chlorpyrifos	16.2	0.1	4	741	-	1	9.3	0.8	4

Treatment	pH			Ammonia Nitrogen (mg/L)			Un-ionized Ammonia (mg/L)		
	Mean	SD	*N*	Mean	SD	*N*	Mean	SD	*N*
Hatchery Tap Water (HTW)	8.17	0.07	4	0.16	0.10	3	0.006	0.003	3
HTW Solvent Control	8.17	0.08	4	0.12	0.16	3	0.004	0.005	3
HTW + 6.25 ppb Chlorpyrifos	8.15	0.08	4	0.13	0.16	3	0.004	0.005	3
HTW + 12.5 ppb Chlorpyrifos	8.15	0.07	4	0.14	0.11	3	0.005	0.003	3
HTW + 25 ppb Chlorpyrifos	8.15	0.08	4	0.12	0.10	3	0.004	0.003	3
HTW + 50 ppb Chlorpyrifos	8.14	0.08	4	0.09	0.11	3	0.003	0.003	3
HTW + 100 ppb Chlorpyrifos	8.16	0.07	4	0.14	0.09	3	0.005	0.003	3
HTW + 200 ppb Chlorpyrifos	8.15	0.08	4	0.07	0.03	3	0.002	0.001	3
HTW + 400 ppb Chlorpyrifos	8.13	0.07	4	0.12	0.07	3	0.004	0.002	3

1. The matrix tested was water from the UC Davis Smelt Hatchery, Tracy, CA (Hardness: 144 mg/L, Alkalinity: 70 mg/L).

Appendix G

P. promelas:

Tests to Determine Effect Concentrations for Select Contaminants

Table G 1-1. Results of a *P. promelas* 7-day test initiated 8/7/08 evaluating the toxicity of copper chloride spiked into laboratory control water and water collected from the UC Davis Smelt Hatchery in Byron, CA.

Treatment	Measured Cu (ppb) *		96 h Survival (%)[1]		7-day Survival (%)[1]		Weight (mg/individual)[1]	
	Total	Dissolved	Mean	SE	Mean	SE	Mean	SE
DIEPAMH	-	-	97.5	2.5	97.5	2.5	0.488	0.021
DIEPAMH @ 900 uS/cm (D900)	-	-	100.0	0.0	100.0	0.0	0.531	0.040
D900 + 15.6 ppb Copper	15.5	14.9	100.0	0.0	100.0	0.0	0.593	0.016
D900 + 31.3 ppb Copper	32.2	31.7	100.0	0.0	100.0	0.0	0.573	0.058
D900 + 62.5 ppb Copper	67.1	60.6	82.5	2.5	67.5	7.5	0.390	0.016
D900 + 125 ppb Copper	129.9	123.3	32.5	6.3	17.5	7.5	0.284	0.086
D900 + 250 ppb Copper	257.9	236.3	0.0	0.0	0.0	0.0	-	-
Hatchery Water @ 900 uS/cm (HW)	2.07	1.72	100.0	0.0	100.0	0.0	0.615	0.035
HW + 15.6 ppb Copper	17.6	16.6	100.0	0.0	100.0	0.0	0.549	0.010
HW + 31.3 ppb Copper	34.3	33.4	100.0	0.0	100.0	0.0	0.512	0.013
HW + 62.5 ppb Copper	69.2	62.3	100.0	0.0	100.0	0.0	0.625	0.029
HW + 125 ppb Copper	132	125	90.0	4.1	75.0	6.5	0.419	0.015
HW + 250 ppb Copper	260	238	35.0	2.9	5.0	2.9	0.380	0.050

*: Copper measurements in DIEPAMH were calculated by subtracting the copper concentrations found in the hatchery water control from the copper concentrations measured in each treatment of the hatchery water dilution series.

1. Highlighted cells indicate significantly reduced survival relative to the solvent control. Data were analyzed using USEPA standard statistical protocols.

Table G 1-2. 96-h and 7-day effect concentrations of copper in a *P. promelas* test initiated on 8/07/08.

Analyte	Endpoint	Matrix	Copper (ppb)					
			NOEC	LOEC	LC10 / EC10		LC50 (survival) / EC25 (weight)	
					Estimate	95% C.I.	Estimate	95% C.I.
Nominal Copper (ppb)	96-hr Survival	DIEPAMH @ 900 uS/cm	31.3	62.5	47	43 - 66	99	87 - 113
		Hatchery Water @ 900 uS/cm	125	250	125	74 - 142	207	180 - 239
	7-day Survival	DIEPAMH @ 900 uS/cm	31.3	62.5	38.9	35 - 48	80.1	70 - 91
		Hatchery Water @ 900 uS/cm	62.5	125	83	72 - 109	154	138 - 172
	7-day Weight	DIEPAMH @ 900 uS/cm	62.5	125	39.7	16 - 44	56.6	48 - 76
		Hatchery Water @ 900 uS/cm	62.5	125	62.36	< 15.6 - 93.4	102	71.1 - 174
Measured Total Copper (ppb)	96-hr Survival	DIEPAMH @ 900 uS/cm	32.2	67.1	49	45 - 63	103	92 - 122
		Hatchery Water @ 900 uS/cm	132	260	132	81 - 150	216	188 - 248
	7-day Survival	DIEPAMH @ 900 uS/cm	32.2	67.1	40	36 - 50	85	67 - 103
		Hatchery Water @ 900 uS/cm	69.2	132	90	79 - 117	162	146 - 180
	7-day Weight	DIEPAMH @ 900 uS/cm	67.1	129.9	41	12 - 45	58	41 - 75
		Hatchery Water @ 900 uS/cm	69.2	132	72.14	< 17.6 - 100.7	109	78.1 - 182
Measured Dissolved Copper (ppb)	96-hr Survival	DIEPAMH @ 900 uS/cm	31.7	60.6	46	43 - 58	96	85 - 115
		Hatchery Water @ 900 uS/cm	125	238	125	74 - 141	200	175 - 228
	7-day Survival	DIEPAMH @ 900 uS/cm	31.7	60.6	39	35 - 47	78	60 - 96
		Hatchery Water @ 900 uS/cm	62.3	125	82	72 - 109	151	136 - 168
	7-day Weight	DIEPAMH @ 900 uS/cm	60.6	123.3	39	12 - 42	53	40 - 67
		Hatchery Water @ 900 uS/cm	62.3	125	62.2	< 1.72 - 238	101.9	70.9 - 171.4

Table G 1-3. Water chemistry during a *P. promelas* 7-day test initiated 8/7/08 evaluating the toxicity of copper chloride spiked into laboratory control water and water collected from the UC Davis Smelt Hatchery in Byron, CA.

Treatment	Laboratory Chemistry							Hardness (mg/L as $CaCO_3$)	Alkalinity (mg/L as $CaCO_3$)	Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)[1]
	EC (uS/cm)	Min Temp (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH				
DIEPAMH	285	24.0	25.3	6.5	8.3	7.64	8.12	84	64	0.00	0.000
DIEPAMH @ 900 uS/cm (D900)	846	24.0	25.5	6.6	8.6	7.56	8.00	128	62	0.00	0.000
D900 + 15.6 ppb Copper	856	24.0	25.4	6.8	8.3	7.60	8.01	-	-	-	-
D900 + 31.3 ppb Copper	842	23.9	25.5	6.4	8.4	7.55	8.04	-	-	-	-
D900 + 62.5 ppb Copper	856	23.9	25.6	6.2	8.4	7.52	8.01	-	-	-	-
D900 + 125 ppb Copper	859	23.9	25.5	6.3	8.4	7.56	7.96	-	-	-	-
D900 + 250 ppb Copper	862	23.9	25.2	7.3	8.2	7.77	7.92	-	-	-	-
Hatchery Water @ 900 uS/cm (HW)	859	23.8	25.6	6.4	8.5	7.60	8.01	120	70	0.05	0.002
HW + 15.6 ppb Copper	857	24.0	25.6	6.5	8.4	7.61	8.07	-	-	-	-
HW + 31.3 ppb Copper	871	23.8	25.6	6.5	8.4	7.61	8.04	-	-	-	-
HW + 62.5 ppb Copper	873	24.0	25.6	6.5	8.4	7.63	8.02	-	-	-	-
HW + 125 ppb Copper	859	23.9	25.7	6.1	8.3	7.57	8.05	-	-	-	-
HW + 250 ppb Copper	859	23.9	25.6	6.7	8.4	7.70	8.09	-	-	-	-

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 G 2-1. Results of a *P. promelas* 7-day test initiated 8/19/2008 evaluating the toxicity of chlorpyrifos in laboratory control water and water collected from the UC Davis Delta Smelt Hatchery in Byron, CA.

Treatment	Measured Chlorpyrifos (ppb)	96-h Survival (%)[1]		7-day Survival (%)[1]		Weight (mg/individual)[1]	
		Mean	SE	Mean	SE	Mean	SE
DIEPAMH	-	100.0	0.0	97.5	2.5	0.400	0.003
DIEPAMH @ 900 uS/cm (D900)	-	100.0	0.0	100.0	0.0	0.462	0.015
D900 Solvent Control	-	100.0	0.0	100.0	0.0	0.443	0.005
D900 + 25 ppb Chlorpyrifos	-	100.0	0.0	100.0	0.0	0.380	0.028
D900 + 50 ppb Chlorpyrifos	-	100.0	0.0	100.0	0.0	0.369	0.023
D900 + 100 ppb Chlorpyrifos	-	100.0	0.0	97.5	2.5	0.314	0.023
D900 + 200 ppb Chlorpyrifos	-	95.0	2.9	90.0	4.1	0.199	0.012
D900 + 400 ppb Chlorpyrifos	-	72.5	2.5	35.0	8.7	0.204	0.098
Hatchery Water @ 900 uS/cm (HW)	-	97.5	2.5	100.0	0.0	0.450	0.019
HW Solvent Control	-	100.0	0.0	100.0	0.0	0.471	0.044
HW + 25 ppb Chlorpyrifos	21.4	100.0	0.0	100.0	0.0	0.417	0.022
HW + 50 ppb Chlorpyrifos	43.2	100.0	0.0	97.5	2.9	0.386	0.026
HW + 100 ppb Chlorpyrifos	82.4	95.0	5.0	95.0	2.5	0.324	0.033
HW + 200 ppb Chlorpyrifos	144	95.0	2.9	77.5	0.0	0.224	0.034
HW + 400 ppb Chlorpyrifos	311	50.0	10.8	27.5	0.0	0.174	0.015

1. Highlighted cells indicate significantly reduced survival relative to the solvent control. Data were analyzed using USEPA standard statistical protocols.

Table G 2-2. 96-h and 7-day effect concentrations of chlorpyrifos in a *P. promelas* test initiated on 8/19/08.

Analyte	Endpoint	Matrix	Chlorpyrifos (ppb)					
			NOEC	LOEC	LC10 / EC10		LC50 (survival) / EC25 (weight)	
					Estimate	95% C.I.	Estimate	95% C.I.
Nominal Chlorpyrifos (ppb)	96-hr Survival	DIEPAMH @ 900 uS/cm	200	400	233	180 - 272	>400	NA
		Hatchery Water @ 900 uS/cm	200	400	219	189 - 244	>400	NA
	7-day Survival	DIEPAMH @ 900 uS/cm	200	400	196	109 - 227	331.0	272 - 442
		Hatchery Water @ 900 uS/cm	200	400	128	53 -253	295.4	258 - 338
	7-day Weight	DIEPAMH @ 900 uS/cm	50	100	8.8	0.4 - 78.4	79.1	39.5 - 133.4
		Hatchery Water @ 900 uS/cm	50	100	32.6	< 25 - 82.8	85.7	32 - 151
Measured Chlorpyrifos (ppb)	96-hr Survival	DIEPAMH @ 900 uS/cm	144	311	170	128 - 204	> 311	NA
		Hatchery Water @ 900 uS/cm	144	311	159	137 - 180	> 311	NA
	7-day Survival	DIEPAMH @ 900 uS/cm	144	311	141	90 - 165	252	202 - 347
		Hatchery Water @ 900 uS/cm	144	311	100	48 - 170	222.1	172 - 299
	7-day Weight	DIEPAMH @ 900 uS/cm	43.2	82.4	7.8	0.5 - 67.4	66	35 - 106
		Hatchery Water @ 900 uS/cm	43.2	82.4	28	< 21.4 - 70.2	144	91 - 267

Table G 2-3. Water chemistry data taken during a *P. promelas* 7-day test initiated 8/19/08 comparing the toxicity of chlorpyrifos between laboratory control water and water from the UC Davis Delta Smelt Hatchery in Byron, CA.

Treatment	Laboratory Chemistry							Hardness (mg/L as $CaCO_3$)	Alkalinity (mg/L as $CaCO_3$)	Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)[1]
	EC (uS/cm)	Min Temp (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH				
DIEPAMH	319	23.6	25.3	7.1	8.3	7.84	8.22	84	58	0.00	0.000
DIEPAMH @ 900 uS/cm (D900)	891	23.5	25.4	7.2	8.3	7.76	8.17	128	60	0.00	0.000
D900 solvent control	903	23.5	25.4	5.9	8.5	7.51	8.11	-	-	-	-
D900 + 25 ppb Chlorpyrifos	902	23.7	25.6	7.2	8.3	7.70	8.14	-	-	-	-
D900 + 50 ppb Chlorpyrifos	894	23.5	25.7	6.5	8.3	7.70	8.14	-	-	-	-
D900 + 100 ppb Chlorpyrifos	885	23.5	25.6	6.2	8.3	7.65	8.16	-	-	-	-
D900 + 200 ppb Chlorpyrifos	882	23.5	25.6	4.9	8.1	7.51	8.66	-	-	-	-
D900 + 400 ppb Chlorpyrifos	888	23.4	25.8	5.0	8.4	7.51	8.15	-	-	-	-
Hatchery Water @ 900 uS/cm (HW)	886	23.5	25.6	7.1	8.1	7.84	8.04	124	72	0.06	0.003
HW Solvent Control	877	23.6	25.8	4.2	8.4	7.31	8.02	-	-	-	-
HW + 25 ppb Chlorpyrifos	885	23.5	25.8	6.7	8.0	7.77	8.02	-	-	-	-
HW + 50 ppb Chlorpyrifos	874	23.6	25.8	6.6	8.2	7.83	8.04	-	-	-	-
HW + 100 ppb Chlorpyrifos	872	23.5	25.9	6.7	8.1	7.71	8.00	-	-	-	-
HW + 200 ppb Chlorpyrifos	875	23.5	26.1	4.2	8.2	7.52	8.03	-	-	-	-
HW + 400 ppb Chlorpyrifos	882	23.5	25.9	3.8	8.3	7.47	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 G 3-1. Results of a *P. promelas* 7-day test initiated 8/19/2008 evaluating the toxicity of esfenvalerate between laboratory control water and water collected from the UC Davis Delta Smelt Hatchery in Byron, CA.

Treatment	Measured Esfenvalerate (ppb)	96-h Survival (%)		7-day Survival (%)		Weight (mg/individual)	
		Mean	SE	Mean	SE	Mean	SE
DIEPAMH	-	100.0	0.0	97.5	2.5	0.465	0.014
DIEPAMH @ 900 uS/cm (D900)	-	100.0	0.0	100.0	0.0	0.411	0.033
D900 solvent control	-	100.0	0.0	100.0	0.0	0.400	0.019
D900 + 0.0625 ppb Esfenvalerate	-	100.0	0.0	100.0	0.0	0.454	0.018
D900 +0.125 ppb Esfenvalerate	-	100.0	0.0	100.0	0.0	0.452	0.028
D900 + 0.250 ppb Esfenvalerate	-	97.5	2.5	97.5	2.5	0.453	0.008
D900 + 0.500 ppb Esfenvalerate	-	100.0	0.0	100.0	0.0	0.512	0.024
D900 +1.000 ppb Esfenvalerate	-	22.5	2.5	5.0	2.9	0.505	0.175
Hatchery Water @ 900 uS/cm (HW)	-	100.0	0.0	100.0	0.0	0.462	0.010
HW Solvent Control	ND	100.0	0.0	97.5	2.5	0.383	0.031
HW + 0.0625 ppb Esfenvalerate	0.049	100.0	0.0	100.0	0.0	0.469	0.018
HW + 0.125 ppb Esfenvalerate	0.150	97.5	2.5	97.5	2.5	0.417	0.025
HW + 0.250 ppb Esfenvalerate	0.208	97.5	2.5	95.0	2.9	0.446	0.025
HW + 0.500 ppb Esfenvalerate	0.500	95.0	2.9	95.0	2.9	0.433	0.034
HW + 1.000 ppb Esfenvalerate	0.920	0.0	0.0	0.0	0.0	-	-

1. Highlighted cells indicate significantly reduced survival relative to the solvent control. Data were analyzed using USEPA standard statistical protocols.

Table G 3-2. 96-h and 7-day effect concentrations of esfenvalerate in a *P. promelas* test initiated on 8/19/08.

Analyte	Endpoint	Matrix	Esfenvalerate (ppb)					
			NOEC	LOEC	LC10 / EC10		LC50 (survival) / EC25 (weight)	
					Estimate	95% C.I.	Estimate	95% C.I.
Nominal Esfenvalerate (ppb)	96-hr Survival	DIEPAMH @ 900 uS/cm	0.500	1.000	0.54	0.52 - 0.55	0.78	0.76 - 0.81
		Hatchery Water @ 900 uS/cm	0.500	1.000	0.52	0.49 - 0.54	0.69	0.67 - 0.71
	7-day Survival	DIEPAMH @ 900 uS/cm	0.500	1.000	0.54	0.52 - 0.54	0.72	0.70 - 0.75
		Hatchery Water @ 900 uS/cm	0.500	1.000	0.52	0.49 - 0.53	0.69	0.67 - 0.71
	7-day Weight	DIEPAMH @ 900 uS/cm	1.000	> 1.000	> 1.000	-	> 1.000	-
		Hatchery Water @ 900 uS/cm	0.500	> 0.500	> 0.500	-	> 0.500	-
Measured Esfenvalerate (ppb)	96-hr Survival	DIEPAMH @ 900 uS/cm	0.500	0.920	0.54	0.52 - 0.55	0.74	0.72 - 0.76
		Hatchery Water @ 900 uS/cm	0.500	0.920	0.52	0.49 - 0.54	0.67	0.65 - 0.68
	7-day Survival	DIEPAMH @ 900 uS/cm	0.500	0.920	0.53	0.52 - 0.54	0.69	0.67 - 0.71
		Hatchery Water @ 900 uS/cm	0.500	0.920	0.52	0.49 - 0.53	0.67	0.65 - 0.68
	7-day Weight	DIEPAMH @ 900 uS/cm	0.500	0.920	> 0.92	-	> 0.92	-
		Hatchery Water @ 900 uS/cm	0.500	> 0.500	> 0.50	-	> 0.50	-

Table G 3-3. Water chemistry data taken during a *P. promelas* 7-day test initiated 8/19/08 comparing the toxicity of esfenvalerate between laboratory control water and water from the UC Davis Delta Smelt Hatchery in Byron, CA.

Treatment	Laboratory Chemistry							Hardness (mg/L as $CaCO_3$)	Alkalinity (mg/L as $CaCO_3$)	Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)[1]
	EC (uS/cm)	Min Temp (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH				
DIEPAMH	293	23.7	26.0	7.1	8.3	7.87	8.15	84	58	0.00	0.000
DIEPAMH @ 900 uS/cm (D900)	879	23.5	26.0	7.3	8.3	7.85	8.11	128	60	0.00	0.000
D900 solvent control	861	23.5	25.8	6.1	8.4	7.67	8.14	-	-	-	-
D900 + 0.0625 ppb Esfenvalerate	884	23.5	25.8	7.1	8.4	7.81	8.13	-	-	-	-
D900 +0.125 ppb Esfenvalerate	878	23.5	25.9	6.0	8.4	7.55	8.15	-	-	-	-
D900 + 0.250 ppb Esfenvalerate	877	23.5	25.9	5.7	8.4	7.61	8.13	-	-	-	-
D900 + 0.500 ppb Esfenvalerate	881	23.5	25.9	4.8	8.4	7.49	8.15	-	-	-	-
D900 +1.000 ppb Esfenvalerate	889	23.5	25.9	5.6	8.4	7.58	8.14	-	-	-	-
Hatchery Water @ 900 uS/cm (HW)	877	23.5	25.9	7.0	8.0	7.87	8.04	124	72	0.06	0.003
HW Solvent Control	884	23.5	25.9	3.8	8.4	7.39	8.00	-	-	-	-
HW + 0.0625 ppb Esfenvalerate	880	23.5	25.9	6.6	8.2	7.85	8.02	-	-	-	-
HW + 0.125 ppb Esfenvalerate	881	23.5	25.9	6.3	8.2	7.09	7.99	-	-	-	-
HW + 0.250 ppb Esfenvalerate	893	23.5	25.9	5.3	8.2	7.56	8.02	-	-	-	-
HW + 0.500 ppb Esfenvalerate	872	23.5	25.9	4.4	8.3	7.46	8.00	-	-	-	-
HW + 1.000 ppb Esfenvalerate	871	23.5	25.7	2.7	8.2	7.29	7.96	-	-	-	-

1: This unionized ammonia reading is based on the ammonia nitrogen measured throughout the duration of the test and upon the water chemistry measured at test initiation.

Table G 4-1. Results of a *P. promelas* 7-day test initiated 9/17/2008 evaluating the toxicity of ammonia chloride spiked into laboratory control water and water collected from the UC Davis Smelt Hatchery in Byron, CA.

Treatment	Measured Ammonia (ppm)		96-h Survival (%)		7-day Survival (%)		Weight (mg/individual)	
	Ammonia Nitrogen	Unionized Ammonia	Mean	SE	Mean	SE	Mean	SE
DIEPAMH	0.0	0.000	100.0	0.0	100.0	0.0	0.420	0.014
DIEPAMH @ 900 uS/cm (D900)	0.0	0.000	100.0	0.0	100.0	0.0	0.431	0.017
D900 + 5 ppm Ammonia	3.5	0.142	100.0	0.0	100.0	0.0	0.441	0.018
D900 + 10 ppm Ammonia	7.3	0.286	100.0	0.0	100.0	0.0	0.432	0.011
D900 + 20 ppm Ammonia	15.0	0.518	100.0	0.0	100.0	0.0	0.460	0.025
D900 + 40 ppm Ammonia	30.8	1.004	45.0	15.5	45.0	15.5	0.420	0.069
D900 + 80 ppm Ammonia	65.2	2.212	0.0	0.0	0.0	0.0	-	-
Hatchery Water @ 900 uS/cm (HW)	0.0	0.001	100.0	0.0	100.0	0.0	0.434	0.018
HW + 5 ppm Ammonia	3.9	0.160	100.0	0.0	100.0	0.0	0.419	0.023
HW + 10 ppm Ammonia	7.7	0.331	100.0	0.0	100.0	0.0	0.412	0.034
HW + 20 ppm Ammonia	15.2	0.629	97.5	2.5	97.5	2.5	0.392	0.017
HW + 40 ppm Ammonia	29.8	1.121	0.0	0.0	0.0	0.0	-	-
HW + 80 ppm Ammonia	60.8	1.697	0.0	0.0	0.0	0.0	-	-

1. Highlighted cells indicate significantly reduced survival relative to the appropriate control. Data were analyzed using USEPA standard statistical protocols.

Table G 4-2. 96-h and 7-day effect concentrations of ammonia in a *P. promelas* test initiated on 9/17/08.

Analyte	Endpoint	Matrix	Ammonia (ppm)					
			NOEC	LOEC	LC10 / EC10		LC50 (survival) / EC25 (weight)	
					Estimate	95% C.I.	Estimate	95% C.I.
Nominal Ammonia	96-hr Survival	DIEPAMH @ 900 uS/cm	20	40	22.7	21 - 28	37.6	28 - 57
		Hatchery Water @ 900 uS/cm	20	40	21.1	20 - 22	27.8	27 - 29
	7-day Survival	DIEPAMH @ 900 uS/cm	20	40	22.7	21 - 28	37.6	28 - 57
		Hatchery Water @ 900 uS/cm	20	40	21.1	20 - 22	27.8	27 - 29
	7-day Weight	DIEPAMH @ 900 uS/cm	40	> 40	> 40	NA	> 40	NA
		Hatchery Water @ 900 uS/cm	20	> 20	> 20	NA	> 20	NA
Measured Ammonia Nitrogen	96-hr Survival	DIEPAMH @ 900 uS/cm	15	30.8	17.1	16 - 21	28.9	22 - 45
		Hatchery Water @ 900 uS/cm	15.2	29.8	16	15 - 16	20.9	20 - 21
	7-day Survival	DIEPAMH @ 900 uS/cm	15	30.8	17.1	16 - 21	28.9	22 - 45
		Hatchery Water @ 900 uS/cm	15.2	29.8	16	15 - 16	20.9	20 - 21
	7-day Weight	DIEPAMH @ 900 uS/cm	30.8	> 30.8	> 30.8	NA	> 30.8	NA
		Hatchery Water @ 900 uS/cm	15.2	> 15.2	> 15.2	NA	> 15.2	NA
Measured Unionized Ammonia	96-hr Survival	DIEPAMH @ 900 uS/cm	0.518	1.004	0.597	0.56 - 0.73	0.954	0.75 - 1.54
		Hatchery Water @ 900 uS/cm	0.629	1.121	0.662	0.63 - 0.68	0.853	0.83 - 0.86
	7-day Survival	DIEPAMH @ 900 uS/cm	0.518	1.004	0.597	0.56 - 0.73	0.954	0.75 - 1.54
		Hatchery Water @ 900 uS/cm	0.629	1.121	0.662	0.63 - 0.68	0.853	0.83 - 0.86
	7-day Weight	DIEPAMH @ 900 uS/cm	1.004	> 1.004	> 1.004	NA	> 1.004	NA
		Hatchery Water @ 900 uS/cm	0.629	> 0.629	> 0.629	NA	> 0.629	NA

Table G 4-3. Water chemistry data taken during a *P. promelas* 7-day test initiated 9/17/08 comparing the toxicity of Ammonia between laboratory control water and water from the UC Davis Delta Smelt Hatchery in Byron, CA.

Treatment	Laboratory Chemistry							Hardness (mg/L as $CaCO_3$)	Alkalinity (mg/L as $CaCO_3$)	Ammonia Nitrogen (mg/L)	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				
DIEPAMH	281	23.6	24.3	7.0	8.5	7.69	8.14	88	60	0.00	0.000
DIEPAMH @ 900 uS/cm (D900)	873	23.5	24.3	6.9	8.5	7.64	8.35	132	60	0.00	0.000
D900 + 5 ppm Ammonia	874	23.4	24.4	6.8	8.5	7.66	7.95	-	-	3.54	0.142
D900 + 10 ppm Ammonia	890	23.8	24.5	7.0	8.6	7.66	7.94	-	-	7.28	0.286
D900 + 20 ppm Ammonia	895	23.6	24.3	6.9	8.5	7.62	7.89	-	-	14.96	0.518
D900 + 40 ppm Ammonia	1142	23.8	24.5	7.2	8.6	7.51	7.85	-	-	30.80	1.004
D900 + 80 ppm Ammonia	1470	23.9	25.0	7.2	8.6	7.66	7.86	-	-	65.20	2.212
Hatchery Water @ 900 uS/cm (HW)	857	23.4	24.4	7.0	8.2	7.77	7.96	128	76	0.03	0.001
HW + 5 ppm Ammonia	857	23.6	24.4	7.0	8.4	7.76	7.96	-	-	3.92	0.160
HW + 10 ppm Ammonia	901	23.6	24.4	7.0	8.3	7.74	7.98	-	-	7.68	0.331
HW + 20 ppm Ammonia	975	23.7	24.4	7.0	8.5	7.67	7.96	-	-	15.20	0.629
HW + 40 ppm Ammonia	1170	23.9	25.0	7.0	8.4	7.78	7.90	-	-	29.80	1.121
HW + 80 ppm Ammonia	1421	24.4	24.4	8.3	8.3	7.79	7.79	-	-	60.80	1.697

1: This unionized ammonia reading is based on the ammonia nitrogen measured throughout the duration of the test and upon the water chemistry measured at test initiation.

Table G 5-1. Results of a *P. promelas* 7-day test initiated 9/24/2008 comparing the toxicity of bifenthrin between laboratory control water and water collected from the UC Davis Smelt Hatchery in Byron, CA.

Treatment	Measured Bifenthrin (ppb)	96-hr Survival (%)		7-day Survival (%)		Weight (mg/individual)	
		Mean	SE	Mean	SE	Mean	SE
DIEPAMH (TAC Control)	-	100.0	0.0	100.0	0.0	0.417	0.011
DIEPAMH @ 900 mS/cm	-	100.0	0.0	100.0	0.0	0.412	0.045
DIEPAMH @ 900 mS/cm Solvent Control	-	100.0	0.0	100.0	0.0	0.434	0.017
DIEPAMH @ 900 uS/cm w/ 0.625 ppb Bifenthrin	-	100.0	0.0	97.5	2.5	0.458	0.032
DIEPAMH @ 900 uS/cm w/ 1.25 ppb Bifenthrin	-	90.0	5.8	82.5	6.3	0.418	0.051
DIEPAMH @ 900 uS/cm w/ 2.5 ppb Bifenthrin	-	35.0	10.4	12.5	6.3	0.576	0.105
DIEPAMH @ 900 uS/cm w/ 5 ppb Bifenthrin	-	2.5	2.5	0.0	0.0	-	-
DIEPAMH @ 900 uS/cm w/ 10 ppb Bifenthrin	-	0.0	0.0	0.0	0.0	-	-
Hatchery Water	-	97.5	2.5	97.5	2.5	0.444	0.022
Hatchery Water Solvent Control	0	100.0	0.0	100.0	0.0	0.474	0.004
Hatchery Water w/ 0.625 ppb Bifenthrin	0.400	92.5	2.5	92.5	2.5	0.414	0.022
Hatchery Water w/ 1.25 ppb Bifenthrin	0.700	65.0	15.0	55.0	11.9	0.670	0.066
Hatchery Water w/ 2.5 ppb Bifenthrin	1.40	22.5	14.4	7.5	7.5	0.743	-
Hatchery Water w/ 5 ppb Bifenthrin	2.86	0.0	0.0	0.0	0.0	-	-
Hatchery Water w/ 10 ppb Bifenthrin	5.00	0.0	0.0	0.0	0.0	-	-

1. Highlighted cells indicate significantly reduced survival relative to the solvent control. Data were analyzed using USEPA standard statistical protocols.

Table G 5-2. 96-h and 7-day effect concentrations of bifenthrin in a *P. promelas* test initiated on 9/24/08.

Analyte	Endpoint	Matrix	Bifenthrin (ppb)					
			NOEC	LOEC	LC10 / EC10		LC50 (survival) / EC25 (weight)	
					Estimate	95% C.I.	Estimate	95% C.I.
Nominal Bifenthrin	96-hr Survival	DIEPAMH @ 900 uS/cm	1.25	2.5	1.25	0.71 - 1.49	2.10	1.70 - 3.02
		Hatchery Water @ 900 uS/cm	0.625	1.25	0.72	0.57 - 1.16	1.66	0.73 - 2.70
	7-day Survival	DIEPAMH @ 900 uS/cm	1.25	2.5	0.91	0.63 - 1.45	1.76	1.58 - 2.04
		Hatchery Water @ 900 uS/cm	0.625	1.25	0.69	0.58 - 0.91	1.39	0.84 - 1.83
	7-day Weight	DIEPAMH @ 900 uS/cm	2.5	> 2.5	> 2.5	-	> 2.5	-
		Hatchery Water @ 900 uS/cm	1.25	> 1.25	> 1.25	-	> 1.25	-
Measured Bifenthrin	96-hr Survival	DIEPAMH @ 900 uS/cm	0.7	1.4	0.70	0.45 - 0.84	1.19	0.94 - 1.53
		Hatchery Water @ 900 uS/cm	0.4	0.7	0.42	0.39 - 0.53	0.92	0.43 - 1.50
	7-day Survival	DIEPAMH @ 900 uS/cm	0.7	1.4	0.54	0.43 - 0.82	1.00	0.87 - 1.12
		Hatchery Water @ 900 uS/cm	0.4	0.7	0.42	0.39 - 0.48	0.76	0.50 - 1.03
	7-day Weight	DIEPAMH @ 900 uS/cm	1.4	>1.4	> 1.4	-	> 1.4	-
		Hatchery Water @ 900 uS/cm	1.4	>1.4	> 1.4	-	> 1.4	-

Table G 5-3. Water chemistry data taken during a *P. promelas* 7-day test initiated 9/24/08 comparing the toxicity of Bifenthrin between laboratory control water and water from the UC Davis Delta Smelt Hatchery in Byron, CA.

Treatment	Laboratory Chemistry						
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH
DIEPAMH	276	23.5	24.4	7.6	8.2	7.86	8.11
DIEPAMH @ 900 uS/cm (D900)	870	23.9	24.5	7.3	8.3	7.77	8.02
D900 solvent control	868	23.9	24.3	6.2	8.3	7.50	8.03
D900 + 0.625 ppb Bifenthrin	874	23.8	24.5	6.9	8.6	7.71	7.99
D900 + 1.25 ppb Bifenthrin	862	23.6	24.5	6.2	8.4	7.64	8.03
D900 + 2.5 ppb Bifenthrin	856	23.7	24.6	5.7	8.3	7.57	8.05
D900 + 5 ppb Bifenthrin	865	24.0	24.4	5.7	8.5	7.58	8.09
D900 + 10 ppb Bifenthrin	863	24.1	24.5	5.7	8.5	7.58	8.02
Hatchery Water @ 900 uS/cm (HW)	878	24.0	24.5	7.4	8.4	7.83	8.04
HW Solvent Control	872	23.7	24.7	4.4	8.6	7.47	8.00
HW + 0.625 ppb Bifenthrin	882	24.0	24.7	7.1	8.5	7.80	8.04
HW + 1.25 ppb Bifenthrin	884	24.0	24.5	5.9	8.5	7.66	8.05
HW + 2.5 ppb Bifenthrin	877	24.2	24.5	5.9	8.6	7.62	8.06
HW + 5 ppb Bifenthrin	873	24.4	24.6	4.8	8.4	7.57	8.04
HW + 10 ppb Bifenthrin	880	24.3	24.5	3.9	8.2	7.44	8.05

Table G 6-1. Results of a *P. promelas* 7-day test initiated 7/07/09 evaluating the toxicity of cyfluthrin in laboratory control water and in water collected from the UC Davis Delta Smelt Hatchery in Byron, CA.

Treatment	Measured Cyfluthrin (ppb)	96-hr Survival (%)[1]		7 Day Survival (%)[1]		Weight (mg/individual)[1]	
		Mean	SE	Mean	SE	Mean	SE
DIEPAMH	-	100.0	0.0	100.0	0.0	0.561	0.019
DIEPAMH @ 900 uS/cm (D900)	-	97.5	2.5	97.5	2.5	0.666	0.034
D900 Solvent Control	-	100.0	0.0	97.5	2.5	0.621	0.037
D900 + 0.125 ppb Cyfluthrin	-	100.0	0.0	97.5	2.5	0.739	0.033
D900 + 0.250 ppb Cyfluthrin	-	100.0	0.0	100.0	0.0	0.649	0.025
D900 + 0.500 ppb Cyfluthrin	-	100.0	0.0	100.0	0.0	0.683	0.009
D900 + 1.000 ppb Cyfluthrin	-	97.5	2.5	87.5	4.8	0.705	0.016
D900 + 2.000 ppb Cyfluthrin	-	2.5	2.5	0.0	0.0	-	-
Hatchery Water @ 900 uS/cm (HW)	-	97.5	2.5	97.5	2.5	0.739	0.037
HW Solvent Control	ND	100.0	0.0	95.0	2.9	0.585	0.041
HW + 0.125 ppb Cyfluthrin	0.076	100.0	0.0	100.0	0.0	0.632	0.031
HW + 0.250 ppb Cyfluthrin	0.129	97.5	2.5	97.5	2.5	0.691	0.004
HW + 0.500 ppb Cyfluthrin	0.200	100.0	0.0	100.0	0.0	0.647	0.020
HW + 1.000 ppb Cyfluthrin	0.645	95.0	2.9	95.0	2.9	0.668	0.068
HW + 2.000 ppb Cyfluthrin	1.110	0.0	0.0	0.0	0.0	-	-

1. Highlighted areas indicate significant reduction in survival or biomass compared to the DIEPAMH control. Data were analyzed using USEPA standard statistical protocols.

Table G 6-2. Nominal and measured 96-h and 7-day effect concentrations of cyfluthrin in a *P. promelas* test initiated on 7/07/09.

Analyte	Endpoint	Matrix	NOEC	LOEC	LC10 / EC10		LC50 (survival) / EC25 (weight)	
					Estimate	95% C.I.	Estimate	95% C.I.
Nominal Cyfluthrin (ppb)	96-hour Survival	DIEPAMHR @ 900 uS/cm	1.00	2.00	1.06	1.00 - 1.09	1.41	1.37 - 1.48
		Hatchery Water	1.00	2.00	1.04	0.98 - 1.09	1.39	1.35 - 1.43
	7-day Survival	DIEPAMHR @ 900 uS/cm	1.00	2.00	0.92	0.59 - 1.14	1.35	1.27 - 1.43
		Hatchery Water	1.00	2.00	1.05	0.99 - 1.09	1.40	1.36 - 1.42
	Weight	DIEPAMHR @ 900 uS/cm	1.00	> 1.00	> 1.00	NA	> 1.00	NA
		Hatchery Water	1.00	> 1.00	> 1.00	NA	> 1.00	NA
Measured Cyfluthrin (ppb)	96-hour Survival	DIEPAMHR @ 900 uS/cm	0.645	1.11	0.67	0.64 - 0.69	0.85	0.83 - 0.88
		Hatchery Water	0.645	1.11	0.66	0.63 - 0.69	0.83	0.81 - 0.85
	7-day Survival	DIEPAMHR @ 900 uS/cm	0.645	1.11	0.56	0.25 - 0.74	0.82	0.77 - 0.85
		Hatchery Water	0.645	1.11	0.67	0.64 - 0.69	0.84	0.82 - 0.85
	Weight	DIEPAMHR @ 900 uS/cm	0.645	> 0.645	> 0.645	-	> 0.645	-
		Hatchery Water	0.645	> 0.645	> 0.645	-	> 0.645	-

Table G 6-3. Water chemistry data taken during a *P. promelas* 7-day test initiated 7/07/09 evaluating the toxicity of Cyfluthrin in laboratory control water and in water collected from the UC Davis Delta Smelt Hatchery in Byron, CA.

Treatment	Laboratory Chemistry						
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH
DIEPAMH	285	23.6	24.6	7.5	8.6	7.61	8.04
DIEPAMH @ 900 uS/cm (D900)	887	23.5	24.7	7.3	8.5	7.78	8.14
D900 Solvent Control	880	23.6	24.8	5.8	8.5	7.53	8.19
D900 + 0.125 ppb Cyfluthrin	881	23.6	24.9	6.7	8.6	7.67	8.19
D900 + 0.250 ppb Cyfluthrin	879	23.5	24.9	6.5	8.5	7.61	8.18
D900 + 0.500 ppb Cyfluthrin	884	23.5	24.9	6.0	8.6	7.55	8.19
D900 + 1.000 ppb Cyfluthrin	884	23.6	25.0	5.7	8.6	7.40	8.20
D900 + 2.000 ppb Cyfluthrin	892	23.5	24.5	5.0	8.6	7.44	8.08
Hatchery Water @ 900 uS/cm (HW)	882	23.8	25.1	7.3	8.6	7.78	8.09
HW Solvent Control	883	23.7	25.2	4.3	8.6	7.43	8.11
HW + 0.125 ppb Cyfluthrin	884	23.5	25.0	7.1	8.6	7.79	8.09
HW + 0.250 ppb Cyfluthrin	884	23.5	25.0	6.7	8.6	7.64	8.06
HW + 0.500 ppb Cyfluthrin	880	23.8	25.0	5.0	8.5	7.48	8.07
HW + 1.000 ppb Cyfluthrin	883	23.8	25.1	4.3	8.6	7.44	8.09
HW + 2.000 ppb Cyfluthrin	887	23.9	24.4	4.4	8.6	7.42	8.09

Table G 7-1. Results of a *P. promelas* 7-day test initiated 7/07/09 evaluating the toxicity of permethrin in laboratory control water and in water collected from the UC Davis Delta Smelt Hatchery in Byron, CA.

Treatment	Measured Total Permethrin (ppb)	96-hr Survival (%)[1]		7 Day Survival (%)[1]		Weight (mg/individual)[1]	
		Mean	SE	Mean	SE	Mean	SE
DIEPAMH	-	100.0	0.0	100.0	0.0	0.759	0.029
DIEPAMH @ 900 uS/cm (D900)	-	97.5	2.5	97.5	2.5	0.664	0.023
D900 Solvent Control	-	100.0	0.0	97.5	2.5	0.754	0.040
D900 + 2 ppb Permethrin	-	100.0	0.0	100.0	0.0	0.757	0.038
D900 + 4 ppb Permethrin	-	100.0	0.0	97.5	2.5	0.844	0.041
D900 + 8 ppb Permethrin	-	72.5	8.5	62.5	11.1	0.915	0.068
D900 + 16 ppb Permethrin	-	0.0	0.0	0.0	0.0	-	-
D900 + 32 ppb Permethrin	-	0.0	0.0	0.0	0.0	-	-
Hatchery Water @ 900 uS/cm (HW)	-	100.0	0.0	100.0	0.0	0.648	0.117
HW Solvent Control	0.00	100.0	0.0	97.5	2.5	0.710	0.042
HW + 2 ppb Permethrin	0.23	97.5	2.5	97.5	2.5	0.773	0.020
HW + 4 ppb Permethrin	0.49	100.0	0.0	100.0	0.0	0.760	0.070
HW + 8 ppb Permethrin	1.20	92.5	4.8	90.0	4.1	0.797	0.034
HW + 16 ppb Permethrin	2.52	0.0	0.0	0.0	0.0	-	-
HW + 32 ppb Permethrin	4.34	0.0	0.0	0.0	0.0	-	-

1. Highlighted areas indicate significant reduction in survival or weight compared to the solvent control. Data were analyzed using USEPA standard statistical protocols.

Table G 7-2. Nominal 96-h and 7-day effect concentrations of permethrin in a *P. promelas* test initiated on 7/07/09.

Analyte	Endpoint	Matrix	Permethrin (ppb)					
					LC10 / EC10		LC50 / EC25	
			NOEC	LOEC	Estimate	95% C.I.	Estimate	95% C.I.
Nominal Permethrin (ppb)	96-hr Survival	DIEPAMH @ 900 uS/cm	4	8	5.2	4.5 - 7.3	10	8.2 - 11.2
		Hatchery Water @ 900 uS/cm	8	16	8.2	4.2 - 8.8	11.1	10.3 - 11.5
	7-day Survival	DIEPAMH @ 900 uS/cm	4	8	4.8	4.1 - 5.8	9.3	6.0 - 10.9
		Hatchery Water @ 900 uS/cm	8	16	8.0	4.3 - 8.7	10.9	10.3 - 11.5
	7-day Weight	DIEPAMH @ 900 uS/cm	8	> 8	> 8	NA	> 8	NA
		Hatchery Water @ 900 uS/cm	8	> 8	> 8	NA	> 8	NA
Measured Permethrin (ppb)	96-hr Survival	DIEPAMH @ 900 uS/cm	0.49	1.20	0.717	0.583 - 1.061	1.545	1.215 - 1.744
		Hatchery Water @ 900 uS/cm	1.20	2.52	1.226	0.627 - 1.338	1.73	1.633 - 1.806
	7-day Survival	DIEPAMH @ 900 uS/cm	0.49	1.20	0.640	0.505 - 0.814	1.428	0.848 - 1.702
		Hatchery Water @ 900 uS/cm	1.20	2.52	1.215	0.679 - 1.344	1.723	1.612 - 1.810
	7-day Weight	DIEPAMH @ 900 uS/cm	1.20	> 1.20	> 1.20	-	> 1.20	-
		Hatchery Water @ 900 uS/cm	1.20	> 1.20	> 1.20	-	> 1.20	-

Table G 7-3. Isomers of permethrin detected in a 7-day chronic *P. promelas* test initiated on 7/07/09.

Nominal Concentration (ppb)	Measured Concentration (ppb)
HW Solvent Control	ND
HW + 2 ppb Permethrin	0.230 (0.084 cis, 0.146 trans)
HW + 4 ppb Permethrin	0.494 (0.176 cis, 0.318 trans)
HW + 8 ppb Permethrin	1.198 (0.420 cis, 0.778 trans)
HW + 16 ppb Permethrin	2.524 (0.934 cis, 1.590 trans)
HW + 32 ppb Permethrin	4.340 (1.540 cis, 2.800 trans)

Table G 7-4. Water chemistry data taken during a *P. promelas* 7-day test initiated 7/07/09 evaluating the toxicity of permethrin in laboratory control water and in water collected from the UC Davis Delta Smelt Hatchery in Byron, CA.

Treatment	Laboratory Chemistry							Hardness (mg/L as $CaCO_3$)	Alkalinity (mg/L as $CaCO_3$)	Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)[1]
	EC (uS/cm)	Min Temp (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH				
DIEPAMH	329	23.5	25.1	6.5	8.4	7.55	8.22	80	56	0.00	0.000
DIEPAMH @ 900 uS/cm (D900)	875	23.6	25.1	6.4	8.3	7.50	8.19	120	64	0.02	0.001
D900 Solvent Control	870	23.6	25.1	3.9	8.5	7.25	8.16	-	-	-	-
D900 + 2 ppb Permethrin	872	23.8	25.1	4.1	8.4	7.28	8.18	-	-	-	-
D900 + 4 ppb Permethrin	869	24.0	25.3	4.0	8.3	7.25	8.39	-	-	-	-
D900 + 8 ppb Permethrin	877	24.1	24.7	3.5	8.5	7.23	7.99	-	-	-	-
D900 + 16 ppb Permethrin	877	24.2	24.7	3.9	8.2	7.25	8.01	-	-	-	-
D900 + 32 ppb Permethrin	875	24.2	24.7	3.9	8.3	7.26	8.02	-	-	-	-
Hatchery Water @ 900 uS/cm (HW)	878	24.0	25.4	6.6	8.6	7.56	8.37	128	66	0.05	0.002
HW Solvent Control	880	24.0	25.3	2.8	8.6	7.25	8.20	-	-	-	-
HW + 2 ppb Permethrin	881	23.8	25.4	6.1	8.5	7.48	8.43	-	-	-	-
HW + 4 ppb Permethrin	879	23.8	25.3	4.2	8.4	7.27	8.12	-	-	-	-
HW + 8 ppb Permethrin	882	24.0	25.5	2.5	8.3	7.23	8.11	-	-	-	-
HW + 16 ppb Permethrin	882	24.3	24.8	1.3	8.2	7.11	8.06	-	-	-	-
HW + 32 ppb Permethrin	880	24.2	24.8	1.3	8.2	7.16	8.05	-	-	-	-

Appendix H

H. azteca:

Tests to Determine Effect Concentrations for Select Contaminants

Table H-1-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 12/12/08 examining the toxicity of cyfluthrin.

Treatment	Measured Cyfluthrin (pptr)	96-hour Survival (%)[1]		10-day Survival (%)[1]		Weight (mg/surviving individual)[1]	
		mean	se	mean	se	mean	se
DIEPAMHR (Method Control)		100	0.0	97	2.8	0.060	0.006
DIEPAMHR @ 900 uS/cm (D900)		100	0.0	100	0.0	0.057	0.008
D900 Solvent Control		100	0.0	100	0.0	0.042	0.005
D900 w/ 0.977 pptr Cyfluthrin	0.7	98	2.5	98	2.5	0.040	0.006
D900 w/ 1.953 pptr Cyfluthrin	1.2	100	0.0	100	0.0	0.053	0.005
D900 w/ 3.906 pptr Cyfluthrin[2]	2.2	20	8.2	15	9.6	0.067	0.003
D900 w/ 7.813 pptr Cyfluthrin[2]	3.7	3	2.5	3	2.5	0.080	-
D900 w/ 15.625 pptr Cyfluthrin	12.7	0	0.0	0	0.0	-	-
DIEPAMHR @ 900 uS/cm + 25 ppb PBO		100	0.0	98	2.3	0.059	0.006
D900 Solvent Control + 25 ppb PBO		100	0.0	100	0.0	0.052	0.006
D900 w/ .098 pptr Cyfluthrin + 25 ppb PBO	ND	98	2.5	98	2.5	0.052	0.010
D900 w/ 0.977 pptr Cyfluthrin + 25 ppb PBO		95	5.0	75	6.5	0.048	0.005
D900 w/ 3.906 pptr Cyfluthrin + 25 ppb PBO		0	0.0	0	0.0	-	-
Hatchery Water		100	0.0	100	0.0	0.064	0.004
Hatchery Water Solvent Control		98	2.5	98	2.5	0.073	0.004
Hatchery Water w/ 0.977 pptr Cyfluthrin	0.8	98	2.5	98	2.5	0.050	0.003
Hatchery Water w/ 1.953 pptr Cyfluthrin	1.5	72	6.0	65	8.4	0.057	0.004
Hatchery Water w/ 3.906 pptr Cyfluthrin[2]	3.1	20	5.8	8	2.5	0.187	0.020
Hatchery Water w/ 7.813 pptr Cyfluthrin	6.4	0	0.0	0	0.0	-	-
Hatchery Water w/ 15.625 pptr Cyfluthrin	15.7	0	0.0	0	0.0	-	-

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.
2. These treatments were excluded from analysis of weight effects because of the difficulty of weighing a small number of surviving animals.

Table H-1-2. Nominal 96-h and 10-day effect concentrations of cyfluthrin in a *H. azteca* test initiated on 12/12/08.

Endpoint	Matrix	Cyfluthrin (pptr)					
		NOEC	LOEC	LC10 / EC10		LC50 / EC25	
				Estimate	95% C.I.	Estimate	95% C.I.
96-hour Survival	DIEPAMHR @ 900 uS/cm	1.953	3.906	2.12	2.04 - 2.21	3.04	2.75 - 3.54
	DIEPAMHR @ 900 + 25 ppb PBO	0.977	3.906	1.07	0.26 - 1.22	2.04	1.77 - 2.16
	Hatchery Water	0.977	1.953	1.30	1.01 - 1.83	2.70	2.25 - 3.17
10-day Survival	DIEPAMHR @ 900 uS/cm	1.953	3.906	2.12	2.05 - 2.20	2.97	2.73 - 3.57
	DIEPAMHR @ 900 + 25 ppb PBO	0.098	0.977	0.34	0.12 - 0.80	1.24	0.95 - 1.63
	Hatchery Water	0.977	1.953	1.22	0.99 - 1.58	2.39	1.95 - 2.83
Weight	DIEPAMHR @ 900 uS/cm	1.953	> 1.953	-	-	-	-
	DIEPAMHR @ 900 + 25 ppb PBO	0.977	> 0.977	-	-	-	-
	Hatchery Water	< 0.977	0.977	0.29	0.16 - 0.66	0.88	0.45 - >3.9

Table H-1-3. Measured 96-h and 10-day effect concentrations of cyfluthrin in a *H. azteca* test initiated on 12/12/08.

Endpoint	Matrix	Cyfluthrin (pptr)					
		NOEC	LOEC	LC10 / EC10		LC50 / EC25	
				Estimate	95% C.I.	Estimate	95% C.I.
96-hour Survival	DIEPAMHR @ 900 uS/cm	1.2	2.2	1.29	1.25 - 1.34	1.77	1.65 - 2.02
	Hatchery Water	0.8	1.5	1.04	0.82 - 1.41	2.12	1.75 - 2.50
10-day Survival	DIEPAMHR @ 900 uS/cm	1.2	2.2	1.29	1.25 - 1.33	1.74	1.61 - 2.04
	Hatchery Water	0.8	1.5	0.98	0.81 - 1.24	1.86	1.50 - 2.22
Weight	DIEPAMHR @ 900 uS/cm	2.2	3.7	-	-	-	-
	Hatchery Water	3.1	> 3.1	-	-	-	-

Table H-1-4. Summary of water chemistry during a *H. azteca* initial screening toxicity test initiated on 12/12/08 examining the toxicity of cyfluthrin.

Treatment	Laboratory Chemistry						
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH
DIEPAMHR (Method Control)	346	20.2	24.6	7.2	8.8	7.72	8.03
DIEPAMHR @ 900 uS/cm (D900)	650	20.2	24.8	7.3	8.5	7.70	7.95
D900 Solvent Control	643	20.5	25.0	5.0	8.2	7.41	8.00
D900 w/ 0.977 pptr Cyfluthrin	919	20.1	24.9	6.9	8.6	7.56	7.92
D900 w/ 1.953 pptr Cyfluthrin	916	19.9	24.8	6.9	8.3	7.69	7.95
D900 w/ 3.906 pptr Cyfluthrin	909	20.3	24.9	7.2	8.6	7.74	7.94
D900 w/ 7.813 pptr Cyfluthrin	890	20.3	24.8	6.6	8.4	7.72	7.94
D900 w/ 15.625 pptr Cyfluthrin	833	21.1	23.3	7.9	8.1	7.92	8.07
DIEPAMHR @ 900 uS/cm + 25 ppb PBO	661	19.9	24.6	6.9	8.3	7.63	8.01
D900 Solvent Control + 25 ppb PBO	660	19.9	24.7	5.0	8.5	7.47	7.98
D900 w/ 0.098 pptr Cyfluthrin + 25 ppb PBO	903	19.9	24.6	7.0	8.5	7.71	7.96
D900 w/ 0.977 pptr Cyfluthrin + 25 ppb PBO	898	19.2	24.7	6.8	8.5	7.78	7.96
D900 w/ 3.906 pptr Cyfluthrin + 25 ppb PBO	836	21.4	23.0	8.0	8.1	7.85	8.04
Hatchery Water	885	20.0	24.4	6.7	8.2	7.96	8.09
Hatchery Water Solvent Control	871	19.9	24.5	3.6	8.2	7.49	8.10
Hatchery Water w/ 0.977 pptr Cyfluthrin	888	20.3	24.5	6.7	8.4	7.89	8.13
Hatchery Water w/ 1.953 pptr Cyfluthrin	894	19.4	24.3	7.0	8.4	7.89	8.14
Hatchery Water w/ 3.906 pptr Cyfluthrin	889	20.3	24.3	7.2	8.5	7.94	8.14
Hatchery Water w/ 7.813 pptr Cyfluthrin	846	21.5	23.6	8.0	8.0	8.06	8.25
Hatchery Water w/ 15.625 pptr Cyfluthrin	845	21.3	23.7	8.0	8.1	8.01	8.24

Table H-2-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 12/30/08 examining the toxicity of diazinon.

Treatment	Measured Diazinon (ppb)	96-hr Survival (%)[1]		10-day Survival (%)[1]		Weight (mg/surviving individual)[1]	
		mean	se	mean	se	mean	se
DIEPAMHR (Method Control)		100	0.0	93	2.5	0.029	0.009
DIEPAMHR @ 900 uS/cm (D900)		100	0.0	100	0.0	0.047	0.002
D900 w/ 0.05% Methanol Control	ND	100	0.0	100	0.0	0.043	0.006
D900 w/ 0.50 ppb Diazinon	0.408	100	0.0	100	0.0	0.040	0.003
D900 w/ 1.00 ppb Diazinon		100	0.0	100	0.0	0.038	0.004
D900 w/ 2.00 ppb Diazinon	2.10	100	0.0	75	9.5	0.019	0.005
D900 w/ 4.00 ppb Diazinon[2]		58	8.5	13	4.8	0.020	0.005
D900 w/ 8.00 ppb Diazinon	9.40	5	3.1	0	0.0	-	-
DIEPAMHR @ 900 uS/cm + 25 ppb PBO		100	0.0	100	0.0	0.035	0.004
D900 w/ 0.05% Methanol Control + 25 ppb PBO		100	0.0	100	0.0	0.036	0.001
D900 w/ 0.40 ppb Diazinon + 25 ppb PBO		100	0.0	100	0.0	0.051	0.005
D900 w/ 4.00 ppb Diazinon + 25 ppb PBO		100	0.0	100	0.0	0.036	0.007
D900 w/ 8.00 ppb Diazinon + 25 ppb PBO		98	2.5	95	2.9	0.021	0.003
Hatchery Water	0.019	98	2.5	98	2.5	0.037	0.004
Hatchery Water w/ 0.05% Methanol Control	ND	100	0.0	100	0.0	0.045	0.003
Hatchery Water w/ 0.5 ppb Diazinon	0.664	100	0.0	100	0.0	0.049	0.003
Hatchery Water w/ 1.00 ppb Diazinon	1.14	98	2.5	98	2.5	0.039	0.002
Hatchery Water w/ 2.00 ppb Diazinon	2.80	100	0.0	95	2.8	0.033	0.002
Hatchery Water w/ 4.00 ppb Diazinon[2]	5.44	68	13.1	23	2.5	0.013	0.004
Hatchery Water w/ 8.00 ppb Diazinon	11.4	5	2.9	0	0.0	-	-

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.
2. These treatments were excluded from analysis of weight effects because of the difficulty of weighing a small number of surviving animals.

Table H-2-2. Nominal 96-h and 10-day effect concentrations of diazinon in a *H. azteca* test initiated on 12/30/08.

Endpoint	Matrix	NOEC	LOEC	LC10	95% C.I.	LC50	95% C.I.
96-hour Survival	DIEPAMHR @ 900 uS/cm	2	4	2.21	1.41 - 2.69	4.44	3.30 - 5.47
	DIEPAMHR @ 900 + 25 ppb PBO	8	> 8	> 8	-	> 8	-
	Hatchery Water	2	4	2.41	2.00 - 3.48	4.90	2.79 - 5.81
10-day Survival	DIEPAMHR @ 900 uS/cm	2	4	1.34	1.15 - 2.35	2.67	2.19 - 3.08
	DIEPAMHR @ 900 + 25 ppb PBO	8	> 8	> 8	-	> 8	-
	Hatchery Water	2	4	2.11	1.95 - 2.24	3.12	3.00 - 3.27
Weight	DIEPAMHR @ 900 uS/cm	1	2	0.93	< 0.50 - 1.39	1.27	0.00 - 1.78
	DIEPAMHR @ 900 + 25 ppb PBO	8	> 8	1.91	0.17 - 6.39	4.7	< 0.40 - 6.80
	Hatchery Water	1	2	1.05	0.55 - 2.02	> 2	-

Table H-2-3. Measured 96-h and 10-day effect concentrations of diazinon in a *H. azteca* test initiated on 12/30/08.

Endpoint	Matrix	Diazinon (ppb)					
		NOEC	LOEC	LC10 / EC10		LC50 / EC25	
				Estimate	95% C.I.	Estimate	95% C.I.
96-hour Survival	DIEPAMHR @ 900 uS/cm	2.1	5.44	2.68	2.46 - 3.26	5.90	4.50 - 6.95
	Hatchery Water	2.8	5.44	3.41	2.98 - 5.00	6.74	4.18 - 8.21
10-day Survival	DIEPAMHR @ 900 uS/cm	1.14	2.1	1.45	1.30 - 1.86	3.08	2.42 - 3.67
	Hatchery Water	2.8	5.44	3.00	2.79 - 3.12	4.31	4.15 - 4.46
Weight	DIEPAMHR @ 900 uS/cm	1.14	2.1	1.03	< 0.41 - 1.52	1.41	< 0.41 - 1.99
	Hatchery Water	1.14	2.8	0.93	0.59 - 1.57	> 2.8	-

Table H-2-4. Summary of water chemistry during a *H. azteca* toxicity test initiated on 12/30/08 examining the toxicity of diazinon.

Treatment	Laboratory Chemistry						
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH
DIEPAMHR (Method Control)	314	20.4	24.1	7.3	8.8	7.83	8.24
DIEPAMHR @ 900 uS/cm (D900)	854	20.3	24.0	7.2	8.9	7.71	8.22
D900 w/ 0.05% Methanol Control	852	20.6	24.2	4.1	8.6	7.47	8.13
D900 w/ 0.50 ppb Diazinon	853	20.5	24.1	6.9	8.8	7.68	8.22
D900 w/ 1.00 ppb Diazinon	856	20.5	23.9	7.1	8.8	7.70	8.11
D900 w/ 2.00 ppb Diazinon	855	20.6	24.1	7.0	8.7	7.67	8.17
D900 w/ 4.00 ppb Diazinon	866	20.6	25.1	4.6	8.6	7.51	8.13
D900 w/ 8.00 ppb Diazinon	810	20.5	24.4	5.1	8.6	7.61	8.16
DIEPAMHR @ 900 uS/cm + 25 ppb PBO	869	20.5	23.9	7.6	8.7	7.73	8.15
D900 w/ 0.05% Methanol Control + 25 ppb PBO	867	21.0	24.0	3.2	8.7	7.49	8.17
D900 w/ 0.40 ppb Diazinon + 25 ppb PBO	864	20.7	23.7	7.0	8.6	7.67	8.15
D900 w/ 4.00 ppb Diazinon + 25 ppb PBO	865	20.5	23.5	4.6	8.5	7.56	8.12
D900 w/ 8.00 ppb Diazinon + 25 ppb PBO	867	20.8	23.9	3.4	8.7	7.45	8.19
Hatchery Water	866	19.3	23.5	7.5	8.7	7.97	8.16
Hatchery Water w/ 0.05% Methanol Control	864	20.2	24.0	3.4	8.7	7.53	8.14
Hatchery Water w/ 0.05 ppb Diazinon	872	20.5	23.7	7.5	8.8	7.89	8.16
Hatchery Water w/ 1.00 ppb Diazinon	863	20.7	23.6	7.1	8.7	7.87	8.13
Hatchery Water w/ 2.00 ppb Diazinon	879	20.7	23.4	4.0	8.8	7.57	8.11
Hatchery Water w/ 4.00 ppb Diazinon	867	20.6	23.5	4.0	8.7	7.56	8.16
Hatchery Water w/ 8.00 ppb Diazinon	822	20.4	23.9	3.9	8.6	7.55	8.13

Table H-3-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 1/14/09 examining the toxicity of bifenthrin.

Treatment	Measured Bifenthrin (pptr)	96-hr Survival (%)[1]		10-day Survival (%)[1]		Weight (mg/surviving individual)[1]	
		mean	se	mean	se	mean	se
DIEPAMHR (Method Control)		100	0.0	100	0.0	0.062	0.006
DIEPAMHR @ 900 uS/cm (D900)		100	0.0	100	0.0	0.078	0.008
D900 w/ 0.05% Methanol Control	ND	98	2.5	98	2.5	0.069	0.004
D900 w/ 1.0 pptr Bifenthrin	0.6	100	0.0	100	0.0	0.062	0.010
D900 w/ 2.0 pptr Bifenthrin		98	2.5	98	2.5	0.053	0.004
D900 w/ 4 pptr Bifenthrin	2.0	100	0.0	100	0.0	0.045	0.003
D900 w/ 8 pptr Bifenthrin[2]		75	5.0	35	6.5	0.028	0.007
D900 w/ 16 pptr Bifenthrin	8.0	8	4.8	0	0.0	-	-
D900 + 25 ppb PBO		100	0.0	100	0.0	0.068	0.006
D900 w/ 0.05% Methanol Control + 25 ppb PBO		100	0.0	100	0.0	0.071	0.008
D900 w/ 0.1 pptr Bifenthrin + 25 ppb PBO		100	0.0	100	0.0	0.085	0.010
D900 w/ 1.0 pptr Bifenthrin + 25 ppb PBO[3]		100	0.0	97	3.3	0.054	0.011
D900 w/ 4 pptr Bifenthrin + 25 ppb PBO[2]		78	4.8	26	10.8	0.083	0.019
Hatchery Water	ND	100	0.0	100	0.0	0.101	0.004
Hatchery Water w/ 0.05% Methanol Control	ND	100	0.0	100	0.0	0.088	0.005
Hatchery Water w/ 1 pptr Bifenthrin	ND	100	0.0	100	0.0	0.089	0.010
Hatchery Water w/ 2 pptr Bifenthrin	ND	100	0.0	100	0.0	0.055	0.010
Hatchery Water w/ 4 pptr Bifenthrin	1.0	100	0.0	98	2.5	0.050	0.002
Hatchery Water w/ 8 pptr Bifenthrin[2]	3.0	90	4.1	33	19.2	0.055	0.016
Hatchery Water w/ 16 pptr Bifenthrin[4]	6.0	13	6.3	3	2.5	0.090	-

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

2. These treatments were excluded from analysis of weight effects because of the difficulty of weighing a small number of surviving animals.

3. This treatment was excluded from analysis of significant differences in survival between treatments because one replicate was missing, and the remaining three replicates did not give sufficient statistical power to detect differences by USEPA methods.

4. This treatment was excluded from analysis of weight because surviving animals were found in only one replicate.

Table H-3-2. Nominal 96-h and 10-day effect concentrations of bifenthrin in a *H. azteca* test initiated on 1/14/09.

Endpoint	Matrix	Bifenthrin (pptr)					
		NOEC	LOEC	LC10 / EC10		LC50 / EC25	
				Estimate	95% C.I.	Estimate	95% C.I.
96-hour Survival	DIEPAMHR @ 900 uS/cm	4	8	5.38	4.56 - 6.27	9.01	9.01 - 11.35
	DIEPAMHR @ 900 + 25 ppb PBO	0.1	4	2.13	1.47 - 4.94	> 4.0	-
	Hatchery Water	8	16	7.87	5.29 - 8.69	11.48	10.57 - 13.28
10-day Survival	DIEPAMHR @ 900 uS/cm	4	8	4.48	4.28 - 4.59	6.88	6.09 - 8.00
	DIEPAMHR @ 900 + 25 ppb PBO	0.1	4	1.18	0.89 - 1.39	2.65	2.03 - 4.61
	Hatchery Water	4	8	4.36	3.98 - 4.99	6.73	5.31 - 11.75
Weight	DIEPAMHR @ 900 uS/cm	2	4	0.86	< 1.0 - 2.25	2.18	< 1.0 - 4.25
	DIEPAMHR @ 900 + 25 ppb PBO	1	> 1	0.33	-	0.77	0.27 - > 4.0
	Hatchery Water	1	2	0.78	< 1.0 - 1.47	1.33	0.50 - 2.05

Table H-3-3. Measured 96-h and 10-day effect concentrations of bifenthrin in a *H. azteca* test initiated on 1/14/09.

Endpoint	Matrix	Bifenthrin (pptr)					
		NOEC	LOEC	LC10 / EC10		LC50 / EC25	
				Estimate	95% C.I.	Estimate	95% C.I.
96-hour Survival	DIEPAMHR @ 900 uS/cm	2	3	2.4	2.2 - 2.6	3.9	3.4 - 4.3
	Hatchery Water	3	6	2.9	1.4 - 3.4	4.3	4.0 - 4.9
10-day Survival	DIEPAMHR @ 900 uS/cm	2	3	2.1	2.1 - 2.2	2.7	2.5 - 3.0
	Hatchery Water	1	3	1.2	1.0 - 1.6	2.3	1.6 - 4.5
Weight	DIEPAMHR @ 900 uS/cm	0.6	2	0.5	< 0.6 - 1.2	1.3	< 0.6 - 2.3
	Hatchery Water	< 1	1	0.2	0.1 - 0.2	0.5	0.4 - 0.7

Table H-3-4. Summary of water chemistry during a *H. azteca* test initiated on 1/14/09 examining the toxicity of bifenthrin.

Treatment	EC (uS/cm)	Min Temp (^{o}C)	Max Temp (^{o}C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH
DIEPAMHR (Method Control)	321	21.8	24.4	7.5	8.6	7.82	8.28
DIEPAMHR @ 900 uS/cm (D900)	846	22.0	24.1	7.6	8.4	7.84	8.16
D900 w/ 0.05% Methanol Control	840	22.0	24.4	4.1	8.6	7.49	8.13
D900 w/ 1 pptr Bifenthrin	856	22.3	24.2	7.6	8.5	7.88	8.11
D900 w/ 2 pptr Bifenthrin	839	22.1	24.5	7.4	8.6	7.81	8.14
D900 w/ 4 pptr Bifenthrin	854	22.2	24.4	7.4	8.6	7.87	8.18
D900 w/ 8 pptr Bifenthrin	841	22.2	24.6	7.7	8.6	7.85	8.18
D900 w/ 16 pptr Bifenthrin	829	22.4	24.4	7.2	8.5	7.82	8.09
Hatchery Water	841	22.3	23.8	7.5	8.7	7.93	8.19
Hatchery Water w/ 0.05% Methanol Control	840	22.3	23.9	3.6	8.7	7.45	8.18
Hatchery Water w/ 1 pptr Bifenthrin	855	22.2	25.5	7.6	8.8	7.92	8.14
Hatchery Water w/ 2 pptr Bifenthrin	853	22.1	24.0	7.6	8.9	5.08	8.20
Hatchery Water w/ 4 pptr Bifenthrin	850	21.9	24.1	7.6	8.7	7.98	8.29
Hatchery Water w/ 8 pptr Bifenthrin	842	22.1	24.0	7.7	8.8	7.92	8.13
Hatchery Water w/ 16 pptr Bifenthrin	839	22.2	24.0	7.5	8.6	7.91	8.15

Table H-4-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 1/15/08 examining the toxicity of chlorpyrifos.

Treatment	Measured Chlorpyrifos (pptr)	96-hr Survival (%)[1]		10-day Survival (%)[1]		Weight (mg/surviving individual)[1]	
		mean	se	mean	se	mean	se
DIEPAMHR (Method Control)		100	0.0	100	0.0	0.073	0.005
DIEPAMHR @ 900 uS/cm (D900)		100	0.0	100	0.0	0.089	0.005
D900 w/ 0.05% Methanol Control	ND	100	0.0	100	0.0	0.071	0.004
D900 w/ 31.25 pptr Chlorpyrifos	14	100	0.0	98	2.5	0.080	0.005
D900 w/ 62.5 pptr Chlorpyrifos		98	2.5	98	2.5	0.092	0.006
D900 w/ 125 pptr Chlorpyrifos[2]	128	68	13.1	31	13.1	0.096	0.002
D900 w/ 250 pptr Chlorpyrifos		3	2.5	0	0.0	-	-
D900 w/ 500 pptr Chlorpyrifos	540	0	0.0	0	0.0	-	-
DIEPAMHR @ 900 uS/cm + 25 ppb PBO		98	2.3	98	2.3	0.081	0.005
D900 w/ 0.05% Methanol Control + 25 ppb PBO		100	0.0	100	0.0	0.074	0.010
D900 12.5 pptr Chlorpyrifos + 25 ppb PBO		100	0.0	100	0.0	0.088	0.004
D900 w/ 125 pptr Chlorpyrifos + 25 ppb PBO		75	5.0	75	5.0	0.078	0.004
D900 w/ 500 pptr Chlorpyrifos + 25 ppb PBO		0	0.0	0	0.0	-	-
Hatchery Water	ND	100	0.0	100	0.0	0.102	0.006
Hatchery Water w/ 0.05% Methanol Control	ND	100	0.0	100	0.0	0.097	0.007
Hatchery Water w/ 31.25 pptr Chlorpyrifos	17	100	0.0	100	0.0	0.101	0.007
Hatchery Water w/ 62.5 pptr Chlorpyrifos	66	100	0.0	100	0.0	0.089	0.008
Hatchery Water w/ 125 pptr Chlorpyrifos[2]	133	59	4.2	21	8.2	0.123	0.011
Hatchery Water w/ 250 pptr Chlorpyrifos	252	0	0.0	0	0.0	-	-
Hatchery Water w/ 500 pptr Chlorpyrifos	420	0	0.0	0	0.0	-	-

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control.
2. These treatments were excluded from analysis of weight effects because of the difficulty of weighing a small number of surviving animals.

Table H-4-2. Nominal 96-h and 10-day effect concentrations of chlorpyrifos in a *H. azteca* test initiated on 1/15/09.

Endpoint	Matrix	Chlorpyrifos (pptr)					
		NOEC	LOEC	LC10 / EC10		LC50 / EC25	
				Estimate	95% C.I.	Estimate	95% C.I.
96-hour Survival	DIEPAMHR @ 900 uS/cm	125	250	75.2	61.4 - 158	152.7	91.2 - 185.8
	DIEPAMHR @ 900 + 25 ppb PBO	12.5	125	32.0	24.7 - 75.0	198.6	178.3 - 235.1
	Hatchery Water	62.5	125	138.8	124.2 - 154.1	138.8	124.2 - 154.1
10-day Survival	DIEPAMHR @ 900 uS/cm	62.5	125	67.6	62.1 - 72.6	102.4	84.3 - 145.6
	DIEPAMHR @ 900 + 25 ppb PBO	12.5	125	32.0	24.7 - 75.0	198.6	178.3 - 235.1
	Hatchery Water	62.5	125	96.7	86.1 - 108.0	101.9	93.2 - 111.4
Weight	DIEPAMHR @ 900 uS/cm	62.5	> 62.5	-	-	-	-
	DIEPAMHR @ 900 + 25 ppb PBO	125	> 125	-	-	-	-
	Hatchery Water	62.5	> 62.5	-	-	-	-

Table H-4-3. Measured 96-h and 10-day effect concentrations of chlorpyrifos in a *H. azteca* test initiated on 1/15/09.

Endpoint	Matrix	Chlorpyrifos (pptr)					
		NOEC	LOEC	LC10 / EC10		LC50 / EC25	
				Estimate	95% C.I.	Estimate	95% C.I.
96-hour Survival	DIEPAMHR @ 900 uS/cm	66	128	77.9	65.4 - 117.6	153.6	95.7 - 183.3
	Hatchery Water	66	133	78.3	75.4 - 83.4	146.6	131.4 - 161.8
10-day Survival	DIEPAMHR @ 900 uS/cm	66	128	71.1	65.6 - 75.9	105.2	88.2 - 141.7
	Hatchery Water	66	133	72.1	70.5 - 73.8	102.6	91.2 - 114.7
Weight	DIEPAMHR @ 900 uS/cm	14	> 14	-	-	-	-
	Hatchery Water	66	> 66	-	-	-	-

Table H-4-4. Summary of water chemistry during a *H. azteca* 10-day test initiated on 1/15/09 examining the toxicity of chlorpyrifos.

Treatment	EC (uS/cm)	Min Temp (^{o}C)	Max Temp (^{o}C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH
DIEPAMHR (Method Control)	323	19.9	23.4	7.9	8.7	7.94	8.28
DIEPAMHR @ 900 uS/cm (D900)	845	20.0	23.5	7.9	8.6	7.86	8.19
D900 w/ 0.05% Methanol Control	848	20.4	23.6	4.2	8.5	7.65	8.17
D900 w/ 31.25 pptr Chlorpyrifos	843	20.4	23.8	7.6	8.7	7.78	8.22
D900 w/ 62.5 pptr Chlorpyrifos	853	22.1	23.8	7.2	8.6	7.82	8.17
D900 w/ 125 pptr Chlorpyrifos	863	20.7	23.8	6.8	8.5	7.74	8.22
D900 w/ 250 pptr Chlorpyrifos	818	22.2	23.7	4.9	8.4	7.63	8.10
D900 w/ 500 pptr Chlorpyrifos	882	22.9	23.5	6.8	8.4	7.72	8.06
Hatchery Water	859	21.9	23.5	7.4	8.8	8.02	8.15
Hatchery Water w/ 0.05% Methanol Control	855	22.4	23.3	3.8	8.8	7.58	8.16
Hatchery Water w/ 31.25 pptr Chlorpyrifos	865	21.7	23.6	7.7	8.7	8.02	8.18
Hatchery Water w/ 62.5 pptr Chlorpyrifos	879	22.1	23.6	7.7	8.9	8.02	8.15
Hatchery Water w/ 125 pptr Chlorpyrifos	863.5	21.9	23.7	7.6	8.9	7.93	8.17
Hatchery Water w/ 250 pptr Chlorpyrifos	822	20.8	23.8	6.9	8.8	7.71	8.10
Hatchery Water w/ 500 pptr Chlorpyrifos	877.5	23.1	23.5	4.5	8.5	7.59	8.15

Table H-5-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 1/21/09 examining the toxicity of permethrin in a variety of matrices.

Treatment	Measured Permethrin (pptr)	96-hr Survival (%)[1]		10-day Survival (%)[1]		Weight (mg/surviving individual)[1]	
		mean	se	mean	se	mean	se
DIEPAMHR (Method Control)		100	0.0	100	0.0	0.054	0.004
DIEPAMHR @ 900 uS/cm (D900)		100	0.0	100	0.0	0.062	0.004
D900 w/ 0.05% Methanol Control	16	100	0.0	100	0.0	0.060	0.005
D900 w/ 6.25 pptr Permethrin	6	100	0.0	98	2.5	0.045	0.003
D900 w/ 12.5 pptr Permethrin		100	0.0	100	0.0	0.041	0.007
D900 w/ 25 pptr Permethrin	19	100	0.0	100	0.0	0.044	0.011
D900 w/ 50 pptr Permethrin		100	0.0	98	2.5	0.051	0.005
D900 w/ 100 pptr Permethrin[2]	90	45	17.1	15	5.0	0.090	0.037
DIEPAMHR @ 900 uS/cm + 25 ppb PBO		98	2.5	98	2.5	0.052	0.012
D900 w/ 0.05% Methanol Control + 25 ppb PBO		100	0.0	100	0.0	0.042	0.005
D900 w/ 0.625 pptr Permethrin + 25 ppb PBO		100	0.0	100	0.0	0.047	0.010
D900 w/ 6.25 pptr Permethrin + 25 ppb PBO		100	0.0	100	0.0	0.035	0.006
D900 w/ 25 pptr Permethrin + 25 ppb PBO[2]		71	5.5	13	4.7	0.125	0.008
Hatchery Water	ND	100	0.0	100	0.0	0.061	0.007
Hatchery Water w/ 0.05% Methanol Control	ND	100	0.0	100	0.0	0.038	0.004
Hatchery Water w/ 6.25 pptr Permethrin	15	100	0.0	100	0.0	0.072	0.007
Hatchery Water w/ 12.5 pptr Permethrin	14	100	0.0	100	0.0	0.062	0.004
Hatchery Water w/ 25 pptr Permethrin	19	100	0.0	94	5.6	0.041	0.008
Hatchery Water w/ 50 pptr Permethrin	40	95	2.9	93	2.5	0.033	0.010
Hatchery Water w/ 100 pptr Permethrin	69	98	2.5	72	6.0	0.038	0.004

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

2. These treatments were excluded from analysis of weight effects because of the difficulty of weighing a small number of surviving animals.

Table H-5-2. Nominal 96-h and 10-day effect concentrations of permethrin in a *H. azteca* test initiated on 1/21/09.

Endpoint	Matrix	Permethrin (pptr)					
		NOEC	LOEC	LC10 / EC10		LC50 / EC25	
				Estimate	95% C.I.	Estimate	95% C.I.
96-hour Survival	DIEPAMHR @ 900 uS/cm	50	100	56.8	53.2 - 66.8	93.9	67.12 - > 100
	DIEPAMHR @ 900 + 25 ppb PBO	6.25	25	10.34	8.31 - 14.01	> 25	-
	Hatchery Water	100	-	> 100	-	>100	-
10-day Survival	DIEPAMHR @ 900 uS/cm	50	100	53.24	50.0 - 55.3	74.6	70.3 - 78.7
	DIEPAMHR @ 900 + 25 ppb PBO	6.25	25	7.4	7.24 - 7.57	14.12	12.71 - 15.69
	Hatchery Water	50	100	54.39	3.31 - 66.72	> 100	-
Weight	DIEPAMHR @ 900 uS/cm	50	> 50	1.30	0.50 - > 100	> 50	-
	DIEPAMHR @ 900 + 25 ppb PBO	6.25	> 6.25	2.35	-	> 6.25	-
	Hatchery Water	100	> 100	16.11	13.63 - 37.16	23.42*	15.38 - 79.28

*: Low weight of solvent control indicates that EC25 estimate may be an artifact of high weights in low concentration treatments.

Table H-5-3. Measured 96-h and 10-day effect concentrations of permethrin in a *H. azteca* test initiated on 1/21/09.

Endpoint	Matrix	Permethrin (pptr)					
		NOEC	LOEC	LC10 / EC10		LC50 / EC25	
				Estimate	95% C.I.	Estimate	95% C.I.
96-hour Survival	DIEPAMHR @ 900 uS/cm	40	90	46.4	43.0 - 56.1	83.6	56.1 - > 90
	Hatchery Water	90	> 90	> 90	-	> 90	-
10-day Survival	DIEPAMHR @ 900 uS/cm	40	90	43.1	40.0 - 45.0	63.9	59.6 - 68.1
	Hatchery Water	40	90	44.1	1.6 - 56.0	> 90	-
Weight	DIEPAMHR @ 900 uS/cm	-	-	-	-	-	-
	Hatchery Water	-	-	-	-	-	-

Note: In this test, weights did not decrease compared to the solvent controls, so any weight decreases relative to the low concentration treatments are probably artifacts and cannot produce reliable estimates of chronic toxicity.

Table H-5-4. Summary of water chemistry during a *H. azteca* 10-day test initiated on 1/21/09 examining the toxicity of permethrin.

Treatment	Laboratory Chemistry						
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH
DIEPAMHR (Method Control)	325	20.2	23.6	8.0	8.6	7.76	8.24
DIEPAMHR @ 900 uS/cm (D900)	821	20.2	23.4	8.2	8.7	7.56	8.40
D900 w/ 0.05% Methanol Control	843	21.0	23.7	4.0	8.8	7.56	8.10
D900 w/ 6.25 pptr Permethrin	831	20.9	23.8	8.0	8.7	7.75	8.13
D900 w/ 12.5 pptr Permethrin	870	21.2	23.7	8.1	8.9	7.78	8.16
D900 w/ 25 pptr Permethrin	836	21.1	23.9	8.0	8.7	7.69	8.20
D900 w/ 50 pptr Permethrin	859	21.2	23.6	7.9	8.8	7.80	8.16
D900 w/ 100 pptr Permethrin	856	21.3	23.8	7.6	8.8	7.71	8.19
Hatchery Water	871	21.8	23.9	7.9	8.9	7.98	8.19
Hatchery Water w/ 0.05% Methanol Control	867	21.3	23.7	3.5	8.7	7.56	8.19
Hatchery Water w/ 6.25 pptr Permethrin	771	21.8	23.9	8.0	8.8	7.98	8.18
Hatchery Water w/ 12.5 pptr Permethrin	881	21.3	23.9	8.0	8.9	7.99	8.15
Hatchery Water w/ 25 pptr Permethrin	857	21.6	23.9	7.7	8.8	7.99	8.18
Hatchery Water w/ 50 pptr Permethrin	864	21.7	23.9	8.0	8.8	7.99	8.14
Hatchery Water w/ 100 pptr Permethrin	861	21.8	24.0	7.9	8.8	7.98	8.18

Table H-6-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 4/10/09 examining the toxicity of ammonia.

Treatment	Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)	96-hour Survival (%)[1]		10-day Survival (%)[1]		Weight (mg/surviving individual)[1]	
			Mean	SE	Mean	SE	Mean	SE
DIEPAMHR (Method Control)	0.02	0.001	98	2.5	95	2.9	0.057	0.006
DIEPAMHR @ 900 µS/cm (D900)	0.03	0.002	100	0.0	98	2.5	0.083	0.003
D900 w/ 6.25 mg/L NH_4Cl	4.70	0.236	100	0.0	100	0.0	0.084	0.004
D900 w/ 12.5 mg/L NH_4Cl	9.05	0.368	100	0.0	100	0.0	0.063	0.012
D900 w/ 25 mg/L NH_4Cl	19.0	0.658	97	2.8	92	8.3	0.066	0.004
D900 w/ 50 mg/L NH_4Cl	37.0	1.010	92	5.3	89	4.6	0.046	0.010
D900 w/ 100 mg/L NH_4Cl	78.0	1.512	69	3.6	49	4.3	0.033	0.005
D900 w/ 200 mg/L NH_4Cl	158.4	2.107	23	9.9	0	0.0	-	-
Hatchery Water	0.1	0.007	100	0.0	100	0.0	0.056	0.005
Hatchery Water w/ 6.25 mg/L NH_4Cl	4.85	0.279	100	0.0	100	0.0	0.072	0.010
Hatchery Water w/ 12.5 mg/L NH_4Cl	10.15	0.554	100	0.0	100	0.0	0.055	0.006
Hatchery Water w/ 25 mg/L NH_4Cl	19.4	0.793	100	0.0	98	2.5	0.071	0.004
Hatchery Water w/ 50 mg/L NH_4Cl	39.2	1.378	92	5.3	84	3.1	0.059	0.009
Hatchery Water w/ 100 mg/L NH_4Cl	76.0	1.702	86	5.5	50	9.6	0.034	0.005
Hatchery Water w/ 200 mg/L NH_4Cl[2]	156.8	2.500	44	5.2	11	7.9	0.125	0.045

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. This treatment was excluded from weight dose-response calculations because of a lack of precision in weighing the few surviving test animals.

Table H-6-2. 96-h and 10-day effect concentrations of ammonia in a *H. azteca* test initiated on 4/10/09.

Analyte	Endpoint	Matrix	NOEC	LOEC	LC10 / EC10		LC50 / EC25	
					Estimate	95% C.I.	Estimate	95% C.I.
Nominal NH_4Cl (mg/L)	96-hour Survival	DIEPAMHR @ 900 uS/cm	50	100	52.3	36.6 - 65.5	130.9	108 - 169
		Hatchery Water	100	200	69.5	46.1 - 88.9	192.9	149 - 304
	10-day Survival	DIEPAMHR @ 900 uS/cm	50	100	57.3	40.5 - 69.1	94.3	81 - 108
		Hatchery Water	25	50	41.6	30.3 - 51.4	94.3	80 - 114
	Weight	DIEPAMHR @ 900 uS/cm	25	50	8.5	3.7 - 21.3	26.7	0.2 - 38.4
		Hatchery Water	200	> 200	51.7	< 6.25 - 65.3	67.9	22.9 - 87.5
Measured Ammonia Nitrogen (mg/L)	96-hour Survival	DIEPAMHR @ 900 uS/cm	37.0	78.0	39.4	27.3 - 49.8	102.2	84 - 133
		Hatchery Water	76.0	156.8	53.9	40.0 - 68.9	149.3	115 - 234
	10-day Survival	DIEPAMHR @ 900 uS/cm	37.0	78.0	42.8	29.5 - 52.3	72.9	62 - 84
		Hatchery Water	19.4	39.2	32.3	23.6 - 39.8	72.9	62 - 88
	Weight	DIEPAMHR @ 900 uS/cm	19.0	37.0	6.3	2.9 - 15.7	20.2	< 4.85 - 28.8
		Hatchery Water	156.8	> 156.8	40.5	< 4.85 - 50.6	52.5	18 - 67
Measured Unionized Ammonia (mg/L)	96-hour Survival	DIEPAMHR @ 900 uS/cm	1.010	1.512	1.025	0.823 - 1.168	1.714	1.542 - 1.976
		Hatchery Water	1.702	2.500	1.513	1.231 - 1.697	2.406	2.138 - 2.99
	10-day Survival	DIEPAMHR @ 900 uS/cm	1.010	1.512	1.113	0.904 - 1.238	1.454	1.331 - 1.564
		Hatchery Water	0.793	1.378	1.151	0.947 - 1.291	1.731	1.591 - 1.904
	Weight	DIEPAMHR @ 900 uS/cm	0.658	1.01	0.292	0.180 - 0.587	0.688	0.107 - 0.876
		Hatchery Water	2.500	> 2.500	1.392	< 0.279 - 1.501	1.516	0.954 - 1.64

Table H-6-3. Water chemistry during a 10-day *H. azteca* water column toxicity test initiated on 4/10/09 examining the toxicity of ammonia.

Treatment	Initial EC (uS/cm)	Temp (°C)		DO (mg/L)		pH	
		Min	Max	Min	Max	Min	Max
DIEPAMHR (Method Control)	338	23.1	23.7	5.8	8.7	7.43	8.18
DIEPAMHR @ 900 µS/cm (D900)	881	23.2	23.8	5.6	8.6	7.40	8.17
D900 w/ 6.25 mg/L NH_4Cl	917	23.2	23.8	5.8	8.7	7.41	8.06
D900 w/ 12.5 mg/L NH_4Cl	956	23.2	23.7	6.2	8.8	7.35	7.97
D900 w/ 25 mg/L NH_4Cl	1039	23.2	24.0	6.4	8.7	7.39	7.89
D900 w/ 50 mg/L NH_4Cl	1214	23.2	23.6	6.5	8.7	7.36	7.80
D900 w/ 100 mg/L NH_4Cl	1567	23.1	24.1	6.6	8.9	7.29	7.64
D900 w/ 200 mg/L NH_4Cl	2157	23.9	23.9	6.7	8.6	7.10	7.49
Hatchery Water	883	23.1	23.5	5.5	8.9	7.57	8.19
Hatchery Water w/ 6.25 mg/L NH_4Cl	910	23.3	23.9	6.6	8.4	7.54	8.12
Hatchery Water w/ 12.5 mg/L NH_4Cl	958	23.2	23.8	6.5	8.5	7.55	8.10
Hatchery Water w/ 25 mg/L NH_4Cl	1046	23.4	23.5	6.3	8.7	7.52	7.98
Hatchery Water w/ 50 mg/L NH_4Cl	1213	23.4	23.7	6.6	8.3	7.50	7.91
Hatchery Water w/ 100 mg/L NH_4Cl	1567	23.7	23.9	6.6	8.4	7.39	7.71
Hatchery Water w/ 200 mg/L NH_4Cl	2204	23.5	23.9	6.5	8.6	7.22	7.57

Treatment	Ammonia Nitrogen (mg/L)		Unionized Ammonia (mg/L)		Hardness (mg/L as $CaCO_3$)	Alkalinity (mg/L as $CaCO_3$)
	Initial	Final	Initial	Final		
DIEPAMHR (Method Control)	0.02	0.71	0.001	0.019	100	56
DIEPAMHR @ 900 µS/cm (D900)	0.03	0.65	0.002	0.010	168	60
D900 w/ 6.25 mg/L NH_4Cl	4.70	1.78	0.236	0.043	-	-
D900 w/ 12.5 mg/L NH_4Cl	9.05	1.79	0.368	0.035	-	-
D900 w/ 25 mg/L NH_4Cl	19.0	1.69	0.658	0.037	-	-
D900 w/ 50 mg/L NH_4Cl	37.0	1.69	1.010	0.030	-	-
D900 w/ 100 mg/L NH_4Cl	78.0	1.67	1.512	0.022	-	-
D900 w/ 200 mg/L NH_4Cl	158.4	-	2.107	-	-	-
Hatchery Water	0.10	0.63	0.007	0.024	148	82
Hatchery Water w/ 6.25 mg/L NH_4Cl	4.85	1.69	0.279	0.063	-	-
Hatchery Water w/ 12.5 mg/L NH_4Cl	10.15	1.66	0.554	0.051	-	-
Hatchery Water w/ 25 mg/L NH_4Cl	19.4	1.55	0.793	0.042	-	-
Hatchery Water w/ 50 mg/L NH_4Cl	39.2	1.67	1.378	0.035	-	-
Hatchery Water w/ 100 mg/L NH_4Cl	76.0	1.74	1.702	0.027	-	-
Hatchery Water w/ 200 mg/L NH_4Cl	156.8	1.60	2.500	0.018	-	-

Table H-7-1. Summary of 10-day *H. azteca* water column toxicity test initiated on 4/10/09 examining the toxicity of copper.

Treatment	Measured Dissolved Copper (ppb)	96-hour Survival (%)[1]		10-day Survival (%)[1]		Weight (mg/surviving individual)[1]	
		Mean	SE	Mean	SE	Mean	SE
DIEPAMHR (Method Control)	-	100	0.0	92	2.6	0.068	0.004
DIEPAMHR @ 900 µS/cm (D900)	ND	100	0.0	100	0.0	0.080	0.003
D900 w/ 125 ppb Copper	122	100	0.0	90	4.1	0.027	0.004
D900 w/ 250 ppb Copper	242	98	2.5	7	7.1	-	-
D900 w/ 500 ppb Copper	491	42	7.8	4	3.6	-	-
D900 w/ 1000 ppb Copper	923	5	3.1	0	0.0	-	-
D900 w/ 2000 ppb Copper	1820	0	0.0	0	0.0	-	-
Hatchery Water	2.80	100	0.0	100	0.0	0.095	0.012
Hatchery Water w/ 125 ppb Copper	124	100	0.0	100	0.0	0.047	0.009
Hatchery Water w/ 250 ppb Copper	-	98	2.5	85	6.5	0.016	0.003
Hatchery Water w/ 500 ppb Copper	504	69	11.2	0	0.0	-	-
Hatchery Water w/ 1000 ppb Copper	-	6	3.6	0	0.0	-	-
Hatchery Water w/ 2000 ppb Copper	1990	0	0.0	0	0.0	-	-

1. Highlighted areas indicate a significant reduction in survival or weight compared to the appropriate control. Data were analyzed using USEPA standard statistical protocols.

Table H-7-2. Nominal 96-h and 10-day effect concentrations of copper in a *H. azteca* test initiated on 4/10/09.

Endpoint	Matrix	Dissolved Copper (ppb)					
		NOEC	LOEC	LC10 / EC10		LC50 (survival) / EC25 (weight)	
				Estimate	95% C.I.	Estimate	95% C.I.
96-hour Survival	DIEPAMHR @ 900 uS/cm	250	500	291	224 - 343	484	422 - 553
	Hatchery Water	500	1000	352	274 - 412	570	500 - 650
10-day Survival	DIEPAMHR @ 900 uS/cm	125	250	125	36 - 140	174	165 - 183
	Hatchery Water	250	500	207	153 - 295	318	293 - 344
Weight	DIEPAMHR @ 900 uS/cm	< 125	125	18	15 - 24	45	38 - 62
	Hatchery Water	< 125	125	24	13 - 57	60	32 - 147

Table H-7-3. Measured 96-h and 10-day effect concentrations of copper in a *H. azteca* test initiated on 4/10/09.

Endpoint	Matrix	Dissolved Copper (ppb)					
		NOEC	LOEC	LC10 / EC10		LC50 (survival) / EC25 (weight)	
				Estimate	95% C.I.	Estimate	95% C.I.
96-hour Survival	DIEPAMHR @ 900 uS/cm	242	491	271	244 - 303	449	377 - 554
	Hatchery Water	504	923	303	230 - 627	613	474 - 737
10-day Survival	DIEPAMHR @ 900 uS/cm	122	242	122	36 - 133	178	165 - 197
	Hatchery Water	242	504	199	152 - 287	343	312 - 369
Weight	DIEPAMHR @ 900 uS/cm	< 122	122	17	15 - 22	44	37 - 57
	Hatchery Water	< 124	124	26	16 - 55	61	36 - 138

Table H-7-4. Water chemistry during a 10-day *H. azteca* water column toxicity test initiated on 4/10/09 examining the toxicity of copper.

Treatment	Laboratory Chemistry							Hardness (mg/L as $CaCO_3$)	Alkalinity (mg/L as $CaCO_3$)	Total Ammonia Nitrogen (mg/L)	Unionized Ammonia (mg/L)
	EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH				
DIEPAMHR (Method Control)	333	22.7	24.0	7.0	8.8	7.73	8.22	100	56	0.02	0.001
DIEPAMHR @ 900 µS/cm (D900)	873	23.1	23.7	7.8	8.6	7.73	8.21	168	60	0.03	0.002
D900 w/ 125 ppb Copper	873	22.8	24.0	7.6	8.7	7.72	8.12	-	-	-	-
D900 w/ 250 ppb Copper	877	22.6	24.0	7.4	8.9	7.88	8.15	-	-	-	-
D900 w/ 500 ppb Copper	872	22.7	24.3	7.7	9.1	7.84	8.12	-	-	-	-
D900 w/ 1000 ppb Copper	854	23.9	23.9	7.7	8.5	7.80	7.94	-	-	-	-
D900 w/ 2000 ppb Copper	861	23.9	23.9	7.9	8.5	7.76	7.91	-	-	-	-
Hatchery Water	870	22.7	24.0	7.9	9.0	7.61	8.16	148	82	0.10	0.006
Hatchery Water w/ 125 ppb Copper	866	22.8	23.8	7.7	8.9	7.82	8.16	-	-	-	-
Hatchery Water w/ 250 ppb Copper	875	22.9	24.1	7.6	8.8	7.89	8.14	-	-	-	-
Hatchery Water w/ 500 ppb Copper	859	22.7	24.2	7.6	8.9	7.94	8.17	-	-	-	-
Hatchery Water w/ 1000 ppb Copper	868	22.6	24.2	7.9	9.0	7.94	8.08	-	-	-	-
Hatchery Water w/ 2000 ppb Copper	855	24.1	24.1	7.9	8.1	7.96	7.96	-	-	-	-

Appendix I

E. affinis:

Tests to Determine Effect Concentrations for Select Contaminants

Table I 1.1. Results of a *E. affinis* 96-hour toxicity test initiated on 3/03/10 examining the toxicity of copper.

Treatment	Measured Dissolved Copper (µg/L)	24-hour Survival (%)[1]	48-hour Survival (%)[1]	72-hour Survival (%)[1]	96-hour Survival (%)[1]
SSEPAMH @ 500	-	100	100	90	90
SSEPAMH @ 900	ND	100	90	70	70
SS900 + 9.375 µg/L Copper	-	100	100	80	80
SS900 + 18.75 µg/L Copper	15.3	100	90	90	90
SS900 + 37.5 µg/L Copper	34.9	100	100	100	30
SS900 + 75 µg/L Copper	72.6	100	100	80	50
SS900 + 150 µg/L Copper	151	100	90	50	30
SS900 + 300 µg/L Copper	305	80	30	10	0

1. Highlighted cells indicate significantly reduced survival relative to appropriate controls. Data were analyzed using USEPA standard statistical protocols.

Table I 1.2. Nominal and measured effect concentrations of copper (µg/L) in a 96-hour *E. affinis* test initiated on 3/03/10.

Analyte	Endpoint	NOEC	LOEC	LC10		LC50		PMSD
				Estimate	95% C.I.	Estimate	95% C.I.	
Nominal Copper (µg/L)	96 hours	150	300	21.6	5.5 - 32.7	75.0	30.2 - 172.3	NA
Measured Copper (µg/L)	96 hours	151	305	18.1	11.3 - 28.5	72.6	27.1 - 174.9	NA

Table I 1.3. Water chemistry during a *E. affinis* 96-hour toxicity test initiated on 3/03/10 examining the toxicity of copper.

Treatment	Temp (°C)		EC (uS/cm)		DO (mg/L)		pH		Hardness (mg/L as CaCO3)	Alkalinity (mg/L as CaCO3)
	Min	Max	Min	Max	Min	Max	Min	Max		
SS900 + 9.375 µg/L Copper	18.7	21	434	473	8.1	9.5	7.9	8.2	120	72
SS900 + 18.75 µg/L Copper	19	20.9	775	851	8.3	9.3	7.8	8.0	156	60
SS900 + 37.5 µg/L Copper	19.2	20.9	780	847	8.1	9.3	7.9	8.1	-	-
SS900 + 75 µg/L Copper	18.8	21.1	783	856	8.3	9.4	7.8	8.1	-	-
SS900 + 150 µg/L Copper	18.8	21.3	779	857	8.2	9.4	7.8	8.1	-	-
SS900 + 300 µg/L Copper	19.1	21.2	780	852	8.1	9.4	7.8	8.1	-	-
SS900 + 9.375 µg/L Copper	19.2	21.3	786	853	8.3	9.4	7.8	8.1	-	-

Table I 2.1. Results of a *E. affinis* 96-hour toxicity test initiated on 3/09/10 examining the toxicity of ammonia.

Treatment	Measured Ammonia Nitrogen (mg/L)	Measured Unionized Ammonia (mg/L)	24-hour Survival (%)[1]	48-hour Survival (%)[1]	72-hour Survival (%)[1]	96-hour Survival (%)[1]
SSEPAMH @ 500 uS/cm	0.0	0.001	100	100	90	70
SSEPAMH @ 900 uS/cm (SS900)	0.0	0.001	100	100	90	70
SS900 + 3.13 mg/L Ammonia	4.0	0.135	100	100	90	30
SS900 + 6.25 mg/L Ammonia	7.0	0.222	100	100	70	20
SS900 + 12.5 mg/L Ammonia	14.7	0.434	80	20	0	0
SS900 + 25 mg/L Ammonia	28	0.767	20	0	0	0
SS900 + 50 mg/L Ammonia	58	1.312	0	0	0	0
SS900 + 100 mg/L Ammonia	114	1.573	0	0	0	0
SS900 + 200 mg/L Ammonia	232	2.681	0	0	0	0

1. Highlighted cells indicate significantly reduced survival relative to appropriate controls. Data were analyzed using USEPA standard statistical protocols.

Table I 2.2. Effect concentrations of ammonia in a 96-hour *E. affinis* test initiated on 3/09/10.

Ammonia Measurement	Endpoint	NOEC	LOEC	LC10		LC50		PMSD
				Estimate	95% C.I.	Estimate	95% C.I.	
Nominal Ammonia Nitrogen (mg/L)	96-hour Survival	6.3	12.5	0.3	0.2 - 3.5	2.5	1.3 - 6.9	NA
Measured Ammonia Nitrogen (mg/L)	96-hour Survival	7.0	14.7	0.3	0.2 - 4.4	3.1	1.5 - 7.8	NA
Measured Unionized Ammonia (mg/L)	96-hour Survival	0.222	0.434	0.023	0.015 - 0.148	0.117	0.076 - 0.250	NA

Table I 2.3. Water chemistry during a *E. affinis* 96-hour toxicity test initiated on 3/09/10 examining the toxicity of copper.

Treatment	Temp (°C)		EC (uS/cm)		DO (mg/L)		pH		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				
SSEPAMH @ 500 uS/cm	19.2	20.7	438	466	7.6	9.2	7.95	8.13	0.0	0.001	120	72
SSEPAMH @ 900 uS/cm (SS900)	19.2	20.5	784	819	7.9	9.5	7.88	8.10	0.0	0.001	156	60
SS900 + 3.13 mg/L Ammonia	19.3	20.9	815	869	8.0	9.5	7.83	8.07	4.0	0.135	-	-
SS900 + 6.25 mg/L Ammonia	19.2	21.0	820	887	7.8	9.5	7.82	8.05	7.0	0.222	-	-
SS900 + 12.5 mg/L Ammonia	19.3	20.8	884	898	8.3	9.5	7.80	8.00	14.7	0.434	-	-
SS900 + 25 mg/L Ammonia	19.4	20.5	980	986	8.4	9.5	7.70	7.93	28.0	0.767	-	-
SS900 + 50 mg/L Ammonia	19.3	20.5	1161	1210	8.6	9.5	7.56	7.85	58.0	1.312	-	-
SS900 + 100 mg/L Ammonia	19.3	20.3	1563	1609	8.5	9.5	7.40	7.71	114.0	1.573	-	-
SS900 + 200 mg/L Ammonia	19.3	20.4	2232	2354	8.6	9.5	7.26	7.58	232.0	2.681	-	-

Table I 3.1. Results of a *E. affinis* 96-hour rangefinding sensitivity test initiated on 3/10/10 examining the toxicity of bifenthrin and cyfluthrin.

Treatment	24-hour Survival (%)[1]	48-hour Survival (%)[1]	72-hour Survival (%)[1]	96-hour Survival (%)[1]
SSEPAMH @ 900 uS/cm, pH 7.9 (SS900)	100	100	75	75
SS900 + 2 pptr Cyfluthrin	100	100	75	50
SS900 + 6 pptr Cyfluthrin	100	100	100	50
SS900 + 18 pptr Cyfluthrin	100	100	50	50
SS900 + 54 pptr Cyfluthrin	50	25	25	25
SS900 + 162 pptr Cyfluthrin	25	25	0	0
SS900 + 486 pptr Cyfluthrin	75	75	0	0
SS900 + 1458 pptr Cyfluthrin	25	0	0	0
SS900 + 4374 pptr Cyfluthrin	0	0	0	0
SS900 + 2 pptr Bifenthrin	100	100	75	50
SS900 + 6 pptr Bifenthrin	100	100	75	75
SS900 + 18 pptr Bifenthrin	100	100	50	50
SS900 + 54 pptr Bifenthrin	100	50	0	0
SS900 + 162 pptr Bifenthrin	100	100	25	25
SS900 + 486 pptr Bifenthrin	25	0	0	0
SS900 + 1458 pptr Bifenthrin	25	0	0	0
SS900 + 4374 pptr Bifenthrin	0	0	0	0

1. Highlighted cells indicate significantly reduced survival relative to the control. Data were analyzed using USEPA standard statistical protocols.

Table I 3.2. Nominal 96-h effect concentrations of cyfluthrin and bifenthrin in a *E. affinis* test initiated on 3/10/10.

Analyte	Endpoint	LC10		LC50	
		Estimate	95% C.I.	Estimate	95% C.I.
Nominal Cyfluthrin (pptr)	96-hr Survival	0.39	0.04 - 37.1	31.30	< 2 - 115.5
Nominal Bifenthrin (pptr)	96-hr Survival	0.93	< 2 - 33.49	26.08	< 2 - 243.6

Table I 3.3. Summary of water chemistry during a *E. affinis* 96-hour rangefinding sensitivity test initiated on 3/10/10 examining the toxicity of bifenthrin and cyfluthrin.

Treatment	Initial EC (uS/cm)	Initial SC (uS/cm)	Temp (°C)		DO (mg/L)		pH		Hardness (mg/L as CaCO3)	Alkalinity (mg/L as CaCO3)
			Min	Max	Min	Max	Min	Max		
SSEPAMH @ 900 uS/cm, pH 7.9 (SS900)	809	901	19.6	20.8	8.5	8.7	8.0	8.0	126	60
SS900 + 2 pptr Cyfluthrin	815	908	19.6	20.8	8.6	8.6	8.0	8.1	-	-
SS900 + 4374 pptr Cyfluthrin	815	910	19.6	20.7	8.5	8.6	8.0	8.1	-	-
SS900 + 2 pptr Bifenthrin	806	899	19.6	20.9	8.5	8.7	7.9	8.1	-	-
SS900 + 4374 pptr Bifenthrin	811	905	19.6	21.0	8.4	8.6	8.0	8.1	-	-

Table I 4.1. Results of a *E. affinis* 96-hour rangefinding sensitivity test initiated on 3/12/10 examining the toxicity of chlorpyrifos, diazinon and permethrin.

Treatment	24-hour Survival (%)[1]	48-hour Survival (%)[1]	72-hour Survival (%)[1]	96-hour Survival (%)[1]
SSEPAMH @ 900 uS/cm, pH 7.9 (SS900)	100	100	75	75
SS900 + 25 pptr Chlorpyrifos	100	100	75	75
SS900 + 75 pptr Chlorpyrifos	100	100	100	50
SS900 + 225 pptr Chlorpyrifos	100	100	100	75
SS900 + 675 pptr Chlorpyrifos	100	100	75	75
SS900 + 2025 pptr Chlorpyrifos	100	75	25	0
SS900 + 6075 pptr Chlorpyrifos	25	0	0	0
SS900 + 50 pptr Diazinon	75	75	75	75
SS900 + 150 pptr Diazinon	100	100	25	25
SS900 + 450 pptr Diazinon	100	100	100	100
SS900 + 1350 pptr Diazinon	100	75	50	50
SS900 + 4050 pptr Diazinon	100	100	100	75
SS900 + 12150 pptr Diazinon	100	100	50	50
SS900 + 30 pptr Permethrin	100	100	100	75
SS900 + 90 pptr Permethrin	100	100	75	75
SS900 + 270 pptr Permethrin	100	100	100	50
SS900 + 810 pptr Permethrin	100	100	100	25
SS900 + 2430 pptr Permethrin	75	25	0	0
SS900 + 7290 pptr Permethrin	0	0	0	0

1. Highlighted cells indicate significantly reduced survival relative to the control. Data were analyzed using USEPA standard statistical protocols.

Table I 4.2. Nominal 96-h effect concentrations of chlorpyrifos, diazinon and permethrin in a *E. affinis* test initiated on 3/12/10.

Analyte	Endpoint	LC10		LC50	
		Estimate	95% C.I.	Estimate	95% C.I.
Nominal Chlorpyrifos (pptr)	96-hr Survival	67.3	< 25 - 1165	1092	< 25 - 1216
Nominal Diazinon (pptr)	96-hr Survival	No Dose - Response			
Nominal Permethrin (pptr)	96-hr Survival	125.2	< 30 - 673.3	467.8	< 30 - 1731

Table I 4.3. Water chemistry during a *E. affinis* 96-hour rangefinding sensitivity test initiated on 3/12/10 examining the toxicity of chlorpyrifos, diazinon and permethrin.

Treatment	Initial EC (uS/cm)	Initial SC (uS/cm)	Temp (°C)		DO (mg/L)		pH		Hardness (mg/L as CaCO3)	Alkalinity (mg/L as CaCO3)
			Min	Max	Min	Max	Min	Max		
SSEPAMH @ 900 uS/cm, pH 7.9 (SS900)	796	882	19.2	20.3	8.7	8.7	7.9	8.1	126	60
SS900 + 6075 pptr Chlorpyrifos	788	872	19.0	19.3	8.5	9.0	8.0	8.1	-	-
SS900 + 12150 pptr Diazinon	783	874	19.1	19.6	8.6	8.9	8.0	8.1	-	-
SS900 + 7290 pptr Permethrin	771	852	19.3	19.8	8.7	9.1	8.0	8.1	-	-

Appendix J

C. dubia:

Tests to Determine Effect Concentrations for Select Contaminants

J.1. 96-hour Acute *C. dubia* Toxicity Tests

Table J 1.1. Results of a 96-hour acute *C. dubia* test initiated on 3/02/10 examining the toxicity of cyfluthrin and bifenthrin.

Treatment	24-hour Survival (%)		48-hour Survival (%)[1]		72-hour Survival (%)		96-hour Survival (%)[1]	
	Mean	SE	Mean	SE	Mean	SE	Mean	SE
SDEPAMH	100	0	100	0	100	0	100	0
SDEPAMH @ 900	100	0	100	0	100	0	100	0
SD900 Solvent Control	100	0	100	0	100	0	100	0
SD900 + 16.625 pptr Cyfluthrin	100	0	100	0	95	5	70	6
SD900 + 31.25 pptr Cyfluthrin	100	0	100	0	70	13	15	10
SD900 + 62.5 pptr Cyfluthrin	100	0	100	0	11	7	0	0
SD900 + 125 pptr Cyfluthrin	100	0	75	5	0	0	0	0
SD900 + 250 pptr Cyfluthrin	100	0	65	10	0	0	0	0
SD900 + 500 pptr Cyfluthrin	100	0	20	14	0	0	0	0
SD900 + 16.625 pptr Bifenthrin	100	0	100	0	95	5	80	8
SD900 + 31.25 pptr Bifenthrin	100	0	90	6	10	10	0	0
SD900 + 62.5 pptr Bifenthrin	100	0	45	13	0	0	0	0
SD900 + 125 pptr Bifenthrin	100	0	45	5	0	0	0	0
SD900 + 250 pptr Bifenthrin	100	0	0	0	0	0	0	0
SD900 + 500 pptr Bifenthrin	100	0	0	0	0	0	0	0

1. Data were analyzed using USEPA standard multiple concentration statistical protocols.

Table J 1.2. Nominal effect concentrations of cyfluthrin and bifenthrin in a *C. dubia* test initiated on 3/02/10.

Pesticide	Endpoint	NOEC	LOEC	LC10		LC50		PMSD
				Estimate	95% C.I.	Estimate	95% C.I.	
Cyfluthrin (pptr)	48 hours	62.5	125	82.5	74.3 - 92.0	315	211 - 508	21.9%
	96 hours	< 16.625	16.625	1.6	0.7 - 4.2	21.0	17.7 - 25.3	19.7%
Bifenthrin (pptr)	48 hours	31.25	62.5	31.3	17.8 - 38.7	57.9	48.1 - 172	20.5%
	96 hours	< 16.625	16.625	3.2	0.1 - 25.6	21.1	18.2 - 23.3	16.3%

Table J 1.3. Water chemistry during a 96-hour acute *C. dubia* toxicity test initiated on 3/02/10 examining the toxicity of cyfluthrin and bifenthrin.

Treatment	EC (uS/cm)		Temp (°C)		DO (mg/L)		pH		Hardness (mg/L as CaCO3)	Alkalinity (mg/L as CaCO3)
	Min	Max	Min	Max	Min	Max	Min	Max		
SDEPAMH	289	307	23.5	26.3	7.3	8.3	7.8	8.2	92	56
SDEPAMH @ 900	851	872	23.9	25.8	7.5	8.5	7.8	8.1	156	60
SD900 Solvent Control	853	861	23.6	25.7	7.5	8.6	7.8	8.1	-	-
SD900 + 16.625 pptr Cyfluthrin	847	861	23.5	25.7	7.4	8.6	7.7	8.1	-	-
SD900 + 31.25 pptr Cyfluthrin	847	868	23.6	25.8	7.3	8.5	7.7	8.1	-	-
SD900 + 62.5 pptr Cyfluthrin	848	862	23.5	25.9	7.4	8.5	7.7	8.1	-	-
SD900 + 125 pptr Cyfluthrin	856	860	23.6	25.9	7.3	8.6	7.8	8.1	-	-
SD900 + 250 pptr Cyfluthrin	856	863	23.6	26.0	7.3	8.4	7.7	8.1	-	-
SD900 + 500 pptr Cyfluthrin	853	861	23.7	25.8	7.2	8.5	7.7	8.1	-	-
SD900 + 16.625 pptr Bifenthrin	860	868	23.6	26.1	7.4	8.3	7.8	8.1	-	-
SD900 + 31.25 pptr Bifenthrin	846	873	23.7	26.6	7.1	8.3	7.7	8.2	-	-
SD900 + 62.5 pptr Bifenthrin	851	869	23.5	26.5	7.3	8.5	7.7	8.1	-	-
SD900 + 125 pptr Bifenthrin	855	870	23.6	26.2	7.4	8.5	7.7	8.1	-	-
SD900 + 250 pptr Bifenthrin	850	857	23.7	25.9	7.5	8.5	7.8	8.1	-	-
SD900 + 500 pptr Bifenthrin	851	858	23.8	26.1	7.5	8.4	7.8	8.1	-	-

Table J 2.1. Results of a 96-hour acute *C. dubia* test initiated on 3/11/10 examining the toxicity of diazinon and chlorpyrifos.

Treatment	24-hour Survival (%)		48-hour Survival (%)[1]		72-hour Survival (%)		96-hour Survival (%)[1]	
	Mean	SE	Mean	SE	Mean	SE	Mean	SE
SDEPAMH	100	0	100	0	100	0	100	0
SDEPAMH @ 900	100	0	100	0	100	0	100	0
SD900 Solvent Control	100	0	100	0	100	0	100	0
SD900 + 50 pptr Diazinon	100	0	100	0	100	0	100	0
SD900 + 100 pptr Diazinon	100	0	100	0	100	0	100	0
SD900 + 200 pptr Diazinon	100	0	95	5	95	5	95	5
SD900 + 400 pptr Diazinon	100	0	65	13	5	5	0	0
SD900 + 800 pptr Diazinon	0	0	0	0	0	0	0	0
SD900 + 1600 pptr Diazinon	0	0	0	0	0	0	0	0
SD900 + 12.5 pptr Chlorpyrifos	100	0	100	0	100	0	100	0
SD900 + 25 pptr Chlorpyrifos	100	0	100	0	100	0	100	0
SD900 + 50 pptr Chlorpyrifos	100	0	100	0	100	0	100	0
SD900 + 100 pptr Chlorpyrifos	80	12	50	24	0	0	0	0
SD900 + 200 pptr Chlorpyrifos	0	0	0	0	0	0	0	0
SD900 + 400 pptr Chlorpyrifos	0	0	0	0	0	0	0	0

1. Data were analyzed using USEPA standard single concentration statistical protocols.

Table J 2.2. Nominal 96-hour effect concentrations of diazinon and chlorpyrifos in a *C. dubia* test initiated on 3/11/10.

Pesticide	Endpoint	NOEC	LOEC	LC10		LC50		PMSD
				Estimate	95% C.I.	Estimate	95% C.I.	
Diazinon (pptr)	48-hr Survival	400	800	224.5	185.3 - 505.3	469.4	319.3 - 589.3	19.10%
	96-hr Survival	200	400	207.4	195.5 - 218.5	277.8	268.9 - 286	10.00%
Chlorpyrifos (pptr)	48-hr Survival	100	200	57.5	52.4 - 125.5	100	60.4 - 157.8	33.60%
	96-hr Survival	50	100	53.6	-	70.8	-	5.50%

Table J 2.3. Water chemistry during a 96-hour acute *C. dubia* toxicity test initiated on 3/11/10 examining the toxicity of diazinon and chlorpyrifos.

Treatment	EC (uS/cm)		Temp (°C)		DO (mg/L)		pH		Hardness (mg/L as CaCO3)	Alkalinity (mg/L as CaCO3)
	Min	Max	Min	Max	Min	Max	Min	Max		
SDEPAMH	289	302	24.0	26.5	6.1	8.4	7.6	8.2	84	60
SDEPAMH @ 900	860	897	23.7	26.2	6.4	8.1	7.5	8.0	126	57
SD900 Solvent Control	869	907	23.8	26.1	6.2	7.9	7.5	8.1	-	-
SD900 + 50 pptr Diazinon	860	895	23.6	26.1	6.5	8.0	7.5	8.1	-	-
SD900 + 100 pptr Diazinon	866	887	23.6	26.2	6.3	8.0	7.6	8.3	-	-
SD900 + 200 pptr Diazinon	859	896	23.7	26.3	6.4	7.9	7.5	8.1	-	-
SD900 + 400 pptr Diazinon	848	888	23.5	27.1	6.3	7.9	7.5	8.1	-	-
SD900 + 800 pptr Diazinon	800	886	24.7	25.4	6.8	7.9	7.7	8.0	-	-
SD900 + 1600 pptr Diazinon	875	887	24.5	25.6	6.8	7.9	7.7	8.0	-	-
SD900 + 12.5 pptr Chlorpyrifos	862	894	23.5	26.1	6.4	8.0	7.6	8.0	-	-
SD900 + 25 pptr Chlorpyrifos	866	891	23.7	26.0	6.3	8.1	7.5	8.0	-	-
SD900 + 50 pptr Chlorpyrifos	865	881	23.9	26.0	6.4	7.8	7.5	8.2	-	-
SD900 + 100 pptr Chlorpyrifos	876	886	23.8	25.8	6.4	7.9	7.5	8.1	-	-
SD900 + 200 pptr Chlorpyrifos	881	883	24.4	25.3	6.8	7.9	7.7	8.0	-	-
SD900 + 400 pptr Chlorpyrifos	878	884	24.4	25.5	6.8	7.9	7.7	8.0	-	-

Table J 3.1. Results of a 96-hour acute *C. dubia* test initiated on 3/16/10 examining the toxicity of permethrin and copper.

Treatment	24-hour Survival (%)		48-hour Survival (%)[1]		72-hour Survival (%)		96-hour Survival (%)[1]	
	Mean	SE	Mean	SE	Mean	SE	Mean	SE
SDEPAMH 030810	100	0	100	0	95	5	95	5
SDEPAMH @ 900	100	0	100	0	100	0	100	0
SD900 Solvent Control	100	0	100	0	100	0	100	0
SD900 + 31.3 pptr Permethrin	100	0	100	0	100	0	100	0
SD900 + 62.5 pptr Permethrin	100	0	100	0	95	5	95	5
SD900 + 125 pptr Permethrin	100	0	100	0	100	0	100	0
SD900 + 250 pptr Permethrin	100	0	100	0	100	0	100	0
SD900 + 500 pptr Permethrin	100	0	95	5	40	14	35	15
SD900 + 1000 pptr Permethrin	55	13	0	0	0	0	0	0
SD900 + 2000 pptr Permethrin	30	10	0	0	0	0	0	0
SD900 + 4000 pptr Permethrin	0	0	0	0	0	0	0	0
SD900 + 18.75 ppb Copper	80	8	70	13	70	13	70	13
SD900 + 37.5 ppb Copper	0	0	0	0	0	0	0	0
SD900 + 75 ppb Copper	0	0	0	0	0	0	0	0
SD900 + 150 ppb Copper	0	0	0	0	0	0	0	0
SD900 + 300 ppb Copper	0	0	0	0	0	0	0	0
SD900 + 600 ppb Copper	0	0	0	0	0	0	0	0
SD900 + 1200 ppb Copper	0	0	0	0	0	0	0	0

1. Data were analyzed using USEPA standard single concentration statistical protocols.

Table J 3.2. Nominal 96-hour effect concentrations of permethrin and copper in a *C. dubia* test initiated on 3/16/10.

Pesticide	Endpoint	NOEC	LOEC	LC10		LC50		PMSD
				Estimate	95% C.I.	Estimate	95% C.I.	
Permethrin (pptr)	48-hr Survival	500	1000	518.6	323.9 - 546.3	694.4	647.7 - 714.8	9.2%
	96-hr Survival	250	500	278.5	256.2 - 311.3	428.7	331.3 - 651.3	22.7%
Copper (ppb)	48-hr Survival	<18.75	18.75	1.6	0.4 - 20.9	22.6	16.4 - 27.1	22.3%
	96-hr Survival	18.75	37.5	1.9	0.3 - 30.6	23.2	17.1 - 28.3	25.9%

Table J 3.3. Water chemistry during a 96-hour acute *C. dubia* toxicity test initiated on 3/16/10 examining the toxicity of permethrin and copper.

Treatment	EC (uS/cm)		Temp (°C)		DO (mg/L)		pH		Hardness (mg/L as CaCO3)	Alkalinity (mg/L as CaCO3)
	Min	Max	Min	Max	Min	Max	Min	Max		
SDEPAMH 030810	294	879	23.5	27.2	6.5	7.9	7.9	8.3	84	60
SDEPAMH @ 900	862	903	23.5	27.1	6.7	8.1	7.8	8.3	126	57
SD900 Solvent Control	853	936	23.5	27.1	6.0	8.1	7.7	8.3	-	-
SD900 + 31.3 pptr Permethrin	847	897	23.3	27.1	6.1	8.0	7.6	8.2	-	-
SD900 + 62.5 pptr Permethrin	850	900	23.4	27.1	6.0	8.0	7.6	8.3	-	-
SD900 + 125 pptr Permethrin	859	901	23.6	27.1	6.1	8.0	7.6	8.2	-	-
SD900 + 250 pptr Permethrin	864	898	23.5	27.1	5.8	8.2	7.7	8.3	-	-
SD900 + 500 pptr Permethrin	858	903	23.6	27.1	6.0	8.1	7.6	8.2	-	-
SD900 + 1000 pptr Permethrin	867	901	23.4	26.1	6.5	7.9	7.9	8.2	-	-
SD900 + 2000 pptr Permethrin	871	899	23.8	27.5	6.2	8.0	7.9	8.2	-	-
SD900 + 4000 pptr Permethrin	847	847	23.5	27.2	6.1	7.9	7.9	8.1	-	-
SD900 + 18.75 ppb Copper	834	888	23.5	27.2	6.7	8.1	7.8	8.2	-	-
SD900 + 37.5 ppb Copper	890	890	23.4	27.0	6.7	7.9	8.0	8.1	-	-
SD900 + 75 ppb Copper	885	885	23.5	26.9	7.0	7.8	8.0	8.1	-	-
SD900 + 150 ppb Copper	894	894	23.5	26.9	7.0	8.0	8.0	8.0	-	-
SD900 + 300 ppb Copper	870	870	23.4	26.9	7.3	7.7	8.0	8.3	-	-
SD900 + 600 ppb Copper	892	892	23.5	26.9	7.3	7.9	8.0	8.0	-	-
SD900 + 1200 ppb Copper	888	888	23.4	27.2	7.3	7.7	7.9	8.1	-	-

Table J 4.1. Results of a 96-hour acute *C. dubia* test initiated on 3/17/10 examining the toxicity of copper.

Treatment	24-hour Survival (%)		48-hour Survival (%)[1]		72-hour Survival (%)		96-hour Survival (%)[1]	
	Mean	SE	Mean	SE	Mean	SE	Mean	SE
SDEPAMH	100	0	100	0	95	5	95	5
SDEPAMH @ 900 µS/cm	100	0	100	0	100	0	100	0
SD900 + 4.69 ppb Copper	100	0	100	0	100	0	100	0
SD900 + 9.38 ppb Copper	100	0	100	0	100	0	100	0
SD900 + 18.75 ppb Copper	100	0	100	0	95	5	95	5
SD900 + 37.5 ppb Copper	0	0	0	0	0	0	0	0
SD900 + 75 ppb Copper	0	0	0	0	0	0	0	0
SD900 + 150 ppb Copper	0	0	0	0	0	0	0	0
SD900 + 300 ppb Copper	0	0	0	0	0	0	0	0
SD900 + 600 ppb Copper	0	0	0	0	0	0	0	0

1. Data were analyzed using USEPA standard single concentration statistical protocols.

Table J 4.2. Nominal 96-hour effect concentrations of copper in a *C. dubia* test initiated on 3/17/10.

Endpoint	NOEC (ppb)	LOEC (ppb)	LC10 (ppb)		LC50 (ppb)		PMSD
			Estimate	95% C.I.	Estimate	95% C.I.	
48-hr Survival	18.75	37.5	20.1	NA	26.6	NA	6.0%
96-hr Survival	18.75	37.5	19.7	13.4 - 20.4	26.3	24.5 - 26.8	12.7%

Table J 4.3. Water chemistry during a 96-hour acute *C. dubia* toxicity test initiated on 3/17/10 examining the toxicity of copper.

Treatment	EC (uS/cm)		Temp (°C)		DO (mg/L)		pH		Hardness (mg/L as CaCO3)	Alkalinity (mg/L as CaCO3)
	Min	Max	Min	Max	Min	Max	Min	Max		
SDEPAMH	834	886	23.7	26.6	7.1	8.2	8.0	8.2	84	60
SDEPAMH @ 900 µS/cm	853	883	24.1	26.2	7.4	8.3	8.0	8.2	126	57
SD900 + 4.69 ppb Copper	868	886	24.2	26.1	7.4	8.0	8.0	8.1	-	-
SD900 + 9.38 ppb Copper	854	893	24.1	26.2	7.4	8.0	8.0	8.1	-	-
SD900 + 18.75 ppb Copper	868	893	24.1	26.2	7.4	8.0	8.0	8.1	-	-
SD900 + 37.5 ppb Copper	864	891	24.0	26.3	7.6	8.1	7.9	8.1	-	-
SD900 + 75 ppb Copper	301	317	24.6	26.8	7.2	8.1	8.0	8.3	-	-
SD900 + 150 ppb Copper	851	874	23.0	26.6	7.0	8.2	7.8	8.1	-	-
SD900 + 300 ppb Copper	856	885	22.5	27.0	7.0	8.2	7.9	8.2	-	-
SD900 + 600 ppb Copper	837	897	22.9	27.1	7.1	8.3	7.9	8.2	-	-

Table J 5.1. Results of a 96-hour acute *C. dubia* test initiated on 4/13/10 examining the toxicity of ammonia.

Treatment	Measured Ammonia Nitrogen (mg/L)	Measured Unionized Ammonia (mg/L)	24-hour Survival (%)		48-hour Survival (%)[1]		72-hour Survival (%)		96-hour Survival (%)[1]	
			Mean	SE	Mean	SE	Mean	SE	Mean	SE
SDEPAMH	0.04	0.003	90	10	90	10	90	10	90	10
SDEPAMH @ 900 µS/cm	0.04	0.003	90	6	90	6	75	13	75	13
SD900 + 3.13 ppm NH3	2.74	0.147	95	5	95	5	75	5	75	5
SD900 + 6.25 ppm NH3	5.18	0.202	100	0	100	0	90	6	90	6
SD900 + 12.5 ppm NH3	9.45	0.236	100	0	100	0	85	5	75	5
SD900 + 25 ppm NH3	19	0.388	95	5	45	13	15	10	0	0
SD900 + 50 ppm NH3	42.4	0.652	0	0	0	0	0	0	0	0
SD900 + 100 ppm NH3	90.4	0.991	0	0	0	0	0	0	0	0
SD900 + 200 ppm NH3	181.6	1.308	0	0	0	0	0	0	0	0
SDEPAMH @ 2675 (High EC)	0.05	0.004	95	5	85	10	85	10	85	10

1. Data were analyzed using USEPA standard single concentration statistical protocols.

Table J 5.2. Nominal and measured 96-hour effect concentrations of ammonia in a *C. dubia* test initiated on 4/13/10.

Analyte	Endpoint	NOEC	LOEC	LC10		LC50		PMSD
				Estimate	95% C.I.	Estimate	95% C.I.	
Nominal Ammonia (mg/L)	48-hr Survival	12.5	25.0	14.3	13.1 - 17.1	24.0	17.8 - 35.1	23.0%
	96-hr Survival	12.5	25.0	12.9	< 3.13 - 13.7	17.4	15.1 - 18.0	31.2%
Measured Ammonia Nitrogen (mg/L)	48-hr Survival	9.45	19.0	10.8	9.8 - 13.0	18.2	13.4 - 28.0	26.3%
	96-hr Survival	9.45	19.0	5.8	< 2.74 - 12.79	12.6	11 - 14	23.1%
Measured Un-ionized Ammonia (mg/L)	48-hr Survival	0.236	0.388	0.263	0.243 - 0.303	0.378	0.312 - 0.514	26.3%
	96-hr Survival	0.236	0.388	0.208	< 0.147 - 0.276	0.295	0.267 - 0.319	23.1%

Table J 5.3. Water chemistry during a 96-hour acute *C. dubia* toxicity test initiated on 4/13/10 examining the toxicity of ammonia.

Treatment	EC (uS/cm)		Temp (°C)		DO (mg/L)		pH		Hardness (mg/L as CaCO3)	Alkalinity (mg/L as CaCO3)
	Min	Max	Min	Max	Min	Max	Min	Max		
SDEPAMH	285.1	3148	24.3	26.2	7.2	8.2	8.1	8.3	92	56
SDEPAMH @ 900 µS/cm	818	928	24.5	26.6	7.4	8.3	7.9	8.2	140	56
SD900 + 3.13 ppm NH3	877	930	24.5	26.4	7.3	8.2	7.9	8.1	-	-
SD900 + 6.25 ppm NH3	935	976	24.1	26.4	7.3	8.5	7.9	8.1	-	-
SD900 + 12.5 ppm NH3	991	1016	23.8	26.6	7.2	8.4	7.7	8.0	-	-
SD900 + 25 ppm NH3	1089	1136	24.0	26.5	7.3	8.3	7.6	7.9	-	-
SD900 + 50 ppm NH3	1317	1320	24.5	25.3	7.4	8.3	7.5	7.8	-	-
SD900 + 100 ppm NH3	1767	1772	24.5	25.4	7.3	8.2	7.4	7.7	-	-
SD900 + 200 ppm NH3	2625	2675	24.5	25.4	7.2	8.2	7.2	7.5	-	-
SDEPAMH @ 2675 (High EC)	2670	2816	24.0	26.8	7.2	8.3	7.7	8.3	-	-

J.2. 7-day Chronic *C. dubia* Toxicity Tests

Table J 6.1. Results of a *C. dubia* 7-day test initiated 10/08/09 evaluating the toxicity of chlorpyrifos in conductivity-adjusted synthetic water and in water collected from the UC Davis Fish Conservation and Culture Laboratory, Skinner Fish Protection Facility, Byron, California.

Treatment	Measured Chlorpyrifos (pptr)	96-hr Survival (%)[1]		7-day Survival (%)[1]		Reproduction (offspring)[1]	
		Mean	SE	Mean	SE	Mean	SE
SSEPAMH (TAC Control)	-	100	0.0	100	0.0	21.4	0.5
SSEPAMH @ 900 uS/cm (SS900)	-	90	10.0	90	10.0	20.9	2.1
SS900 Solvent Control	-	100	0.0	100	0.0	21.2	0.4
SS900 + 12.5 pptr Chlorpyrifos	-	100	0.0	100	0.0	22.1	0.7
SS900 + 25 pptr Chlorpyrifos	-	100	0.0	100	0.0	21.3	0.4
SS900 + 50 pptr Chlorpyrifos	-	100	0.0	100	0.0	20.8	1.0
SS900 + 100 pptr Chlorpyrifos	-	20	13.3	0	0.0	2.4	0.7
SS900 + 200 pptr Chlorpyrifos	-	0	0.0	0	0.0	0.0	0.0
Hatchery Water @ 900 uS/cm (HW900)	-	100	0.0	100	0.0	22.9	0.4
HW900 Solvent Control	ND	100	0.0	100	0.0	21.3	0.9
HW900 + 12.5 pptr Chlorpyrifos	ND	100	0.0	100	0.0	22.3	0.9
HW900 + 25 pptr Chlorpyrifos	11	80	13.3	80	13.3	17.4	2.9
HW900 + 50 pptr Chlorpyrifos	20	100	0.0	100	0.0	20.3	1.1
HW900 + 100 pptr Chlorpyrifos	28	10	10.0	0	0.0	2.8	0.6
HW900 + 200 pptr Chlorpyrifos	56	0	0.0	0	0.0	0.0	0.0

1. Highlighted cells indicate statistically significant reductions in survival or reproduction compared to the laboratory control. Data were analyzed using USEPA standard statistical protocols.

Table J 6.2. Nominal 7-day effect concentrations of chlorpyrifos in a *C. dubia* test initiated on 10/08/09.

Endpoint	Matrix	Chlorpyrifos (pptr)						
		NOEC	LOEC	LC10/EC10		LC50/EC25		
				Estimate	95% C.I.	Estimate	95% C.I.	PMSD
96-hr Survival	SSEPAMH @ 900 uS/cm	50	100	54.6	53.6 - 56.2	77.2	70.8 - 89.1	-
	Hatchery Water @ 900 uS/cm	50	100	50.0	17.7 - 54.0	70.8	64.9 - 79.4	-
7-day Survival	SSEPAMH @ 900 uS/cm	50	100	53.6	-	70.8	-	-
	Hatchery Water @ 900 uS/cm	50	100	50.0	17.7 - 53.6	68.1	64.9 - 70.8	-
7-day Reproduction	SSEPAMH @ 900 uS/cm	50	100	52.4	35.1 - 54.2	59.3	54.5 - 61.3	9.5%
	Hatchery Water @ 900 uS/cm	50	100	17.6	9.0 - 51.4	53.8	21.5 - 58.5	20.5%

Table J 6.3. Measured 7-day effect concentrations of chlorpyrifos in a *C. dubia* test initiated on 10/08/09.

Endpoint	Matrix	Chlorpyrifos (pptr)						
		NOEC	LOEC	LC10/EC10		LC50/EC25		
				Estimate	95% C.I.	Estimate	95% C.I.	PMSD
96-hour Survival	SSEPAMH @ 900 uS/cm	20	28	20.9	20.7 - 21.2	24.7	23.7 - 26.5	-
	Hatchery Water @ 900 uS/cm	20	28	20.0	1.7 - 20.8	23.7	22.7 - 25.0	-
7-day Survival	SSEPAMH @ 900 uS/cm	20	28	20.7	-	23.7	-	-
	Hatchery Water @ 900 uS/cm	20	28	20.0	1.70 - 20.7	23.2	22.4 - 23.7	-
7-day Reproduction	SSEPAMH @ 900 uS/cm	20	28	20.6	18.4 - 20.8	21.9	21.2 - 22.1	9.5%
	Hatchery Water @ 900 uS/cm	20	28	7.7	1.52 - 20.8	21.3	9.13 - 22.1	23.5%

Table J 6.4. Water chemistry data taken during a *C. dubia* 7-day test initiated 10/08/09 evaluating the toxicity of chlorpyrifos in conductivity-adjusted synthetic water and in water collected from the UC Davis Fish Conservation and Culture Laboratory, Skinner Fish Protection Facility, Byron, California.

Treatment	Laboratory Chemistry							Hardness (mg/L as $CaCO_3$)	Alkalinity (mg/L as $CaCO_3$)
	Initial EC (uS/cm)	Min Temp (^oC)	Max Temp (^oC)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH		
SSEPAMH (TAC Control)	184	23.9	27.4	7.4	8.4	7.89	8.16	68	56
SSEPAMH @ 900 uS/cm (SS900)	883	23.8	25.5	7.3	9.1	7.82	8.04	128	56
SS900 Solvent Control	884	23.9	25.5	6.8	8.6	7.78	8.05	-	-
SS900 + 12.5 pptr Chlorpyrifos	881	23.8	25.6	6.8	8.6	7.72	8.04	-	-
SS900 + 25 pptr Chlorpyrifos	881	23.8	25.7	6.9	8.6	7.76	8.03	-	-
SS900 + 50 pptr Chlorpyrifos	877	23.7	26.0	6.8	8.6	7.80	8.01	-	-
SS900 + 100 pptr Chlorpyrifos	882	23.8	25.1	6.9	8.6	7.78	8.02	-	-
SS900 + 200 pptr Chlorpyrifos	876	23.7	24.9	7.5	8.5	7.82	7.99	-	-
Hatchery Water @ 900 uS/cm (HW900)	877	23.7	26.0	7.0	8.6	7.84	8.25	128	78
HW900 Solvent Control	884	23.3	26.0	5.5	8.6	7.69	8.25	-	-
HW900 + 12.5 pptr Chlorpyrifos	879	23.3	25.9	5.6	8.6	7.69	8.25	-	-
HW900 + 25 pptr Chlorpyrifos	864	23.4	25.8	5.7	8.6	7.69	8.24	-	-
HW900 + 50 pptr Chlorpyrifos	882	23.3	26.0	5.8	8.6	7.68	8.23	-	-
HW900 + 100 pptr Chlorpyrifos	882	23.6	25.2	6.8	8.6	7.71	8.24	-	-
HW900 + 200 pptr Chlorpyrifos	879	23.6	24.9	7.4	8.4	7.87	8.15	-	-

Table J 7.1. Results of a *C. dubia* 7-day test initiated 10/22/09 evaluating the toxicity of diazinon in conductivity-adjusted synthetic water and in water collected from the UC Davis Fish Conservation and Culture Laboratory, Skinner Fish Protection Facility, Byron, California.

Treatment	Measured Diazinon (pptr)	96-hour Survival (%)[1]		7-Day Survival (%)[1]		Reproduction (offspring)[1]	
		Mean	SE	Mean	SE	Mean	SE
SSEPAMH (TAC Control)	-	100	0.00	100	0.00	26.3	0.58
SSEPAMH @ 900 uS/cm (SS900)	-	100	0.00	100	0.00	27.1	0.82
SS900 Solvent Control	-	100	0.00	100	0.00	26.5	0.65
SS900 + 62.5 pptr Diazinon	-	100	0.00	100	0.00	25.8	0.83
SS900 + 125 pptr Diazinon	-	100	0.00	100	0.00	26.0	0.87
SS900 + 250 pptr Diazinon	-	100	0.00	0	0.00	15.6	1.47
SS900 + 500 pptr Diazinon	-	0	0.00	0	0.00	0.0	0.00
SS900 + 1000 pptr Diazinon	-	0	0.00	0	0.00	0.0	0.00
Hatchery Water @ 900 uS/cm (HW900)	-	100	0.00	100	0.00	16.4	1.70
HW900 Solvent Control	ND	100	0.00	100	0.00	13.6	1.96
HW900 + 62.5 pptr Diazinon	57	80	13.33	80	0.13	11.5	2.00
HW900 + 125 pptr Diazinon	123	100	0.00	90	0.10	12.8	1.92
HW900 + 250 pptr Diazinon	228	100	0.00	10	0.10	8.8	1.61
HW900 + 500 pptr Diazinon	560	0	0.00	0	0.00	0.0	0.00
HW900 + 1000 pptr Diazinon	1100	0	0.00	0	0.00	0.0	0.00

1. Highlighted cells indicate statistically significant reductions in survival or reproduction compared to the laboratory control. Data were analyzed using USEPA standard statistical protocols.

Table J 7.2. Nominal effect concentrations of diazinon in a 7-day chronic *C. dubia* test initiated on 10/22/09.

Endpoint	Matrix	Diazinon (pptr)						
		NOEC	LOEC	LC10/EC10		LC50/EC25		PMSD
				Estimate	95% C.I.	Estimate	95% C.I.	
96-hour Survival	SSEPAMH @ 900 uS/cm	250	500	268	-	353.6	-	NA
	Hatchery Water @ 900 uS/cm	250	500	256.3	11.1 - 268	345	329.9 - 353.6	NA
7-day Survival	SSEPAMH @ 900 uS/cm	125	250	134.0	-	176.8	142 - 220.1	NA
	Hatchery Water @ 900 uS/cm	125	250	14.9	2.99-134	172.7	150.4 - 198.3	N/A
7-day Reproduction	SSEPAMH @ 900 uS/cm	125	250	145.3	126.5-155.1	189.5	170.9-222.1	10.01%
	Hatchery Water @ 900 uS/cm	250	500	4.0	1.61-142.9	53.85	9.98-237.9	33.35%

Table J 7.3. Measured effect concentrations of diazinon in a 7-day chronic C. dubia test initiated on 10/22/09.

Endpoint	Matrix	Diazinon (pptr)						
		NOEC	LOEC	LC10/EC10		LC50/EC25		PMSD
				Estimate	95% C.I.	Estimate	95% C.I.	
96-hour Survival	SSEPAMH @ 900 uS/cm	228	560	249.5	-	357.4	-	NA
	Hatchery Water @ 900 uS/cm	228	560	235.4	10.4 - 249.5	346.1	326.7 - 357.4	NA
7-day Survival	SSEPAMH @ 900 uS/cm	123	228	130.8	-	167.5	-	NA
	Hatchery Water @ 900 uS/cm	123	228	14.0	2.87 - 130.8	164.1	146.8 - 185.7	NA
7-day Reproduction	SSEPAMH @ 900 uS/cm	123	228	139.1	123.1 - 148.1	176.5	160.3 - 208	11.42%
	Hatchery Water @ 900 uS/cm	228	560	44.1	2.41 - 228	176.2	20.5 - 264.9	41.62%

Table J 7.4. Water chemistry data taken during a *C. dubia* 7-day test initiated 10/22/09 evaluating the toxicity of diazinon in conductivity-adjusted synthetic water and in water collected from the UC Davis Fish Conservation and Culture Laboratory, Skinner Fish Protection Facility, Byron, California.

Treatment	Initial EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH	Hardness (mg/L as $CaCO_3$)	Alkalinity (mg/L as $CaCO_3$)
SSEPAMH (TAC Control)	228	22.9	24.4	7.3	8.4	7.76	8.18	80	58
SSEPAMH @ 900 uS/cm (SS900)	865	22.9	24.3	7.5	8.6	7.80	8.09	-	-
SS900 Solvent Control	871	22.7	24.4	6.9	8.6	7.72	8.10	-	-
SS900 + 62.5 pptr Diazinon	880	22.8	24.5	7.0	8.5	7.70	8.09	-	-
SS900 + 125 pptr Diazinon	878	22.7	24.5	7.1	8.6	7.72	8.08	-	-
SS900 + 250 pptr Diazinon	901	22.6	25.7	7.0	8.6	7.78	8.12	-	-
SS900 + 500 pptr Diazinon	876	23.2	24.6	7.6	8.4	7.80	8.08	-	-
SS900 + 1000 pptr Diazinon	886	23.8	24.9	7.6	8.6	7.84	8.09	-	-
Hatchery Water @ 900 uS/cm (HW900)	863	23.1	24.7	7.3	8.6	7.84	8.23	144	79
HW900 Solvent Control	889	23.0	24.9	4.1	8.6	7.50	8.22	-	-
HW900 + 62.5 pptr Diazinon	865	22.9	24.6	4.1	8.6	7.50	8.24	-	-
HW900 + 125 pptr Diazinon	863	22.9	24.6	4.3	8.6	7.52	8.21	-	-
HW900 + 250 pptr Diazinon	876	22.8	24.6	4.7	8.6	7.52	8.19	-	-
HW900 + 500 pptr Diazinon	866	23.6	24.6	7.3	8.5	7.85	8.17	-	-
HW900 + 1000 pptr Diazinon	879	23.8	24.6	7.3	8.3	7.85	8.18	-	-

Table J 8.1. Results of a *C. dubia* 7-day test initiated 12/02/09 evaluating the toxicity of permethrin in both conductivity-adjusted synthetic water and water from the UC Davis Fish Conservation and Culture Laboratory, Skinner Fish Protection Facility, Byron, California.

Treatment	96-hr Survival (%)[1]		7-day Survival (%)[1]		Reproduction (offspring)[1]	
	Mean	SE	Mean	SE	Mean	SE
SSEPAMH	100	0.0	100	0.0	10.1	2.26
SSEPAMH @ 900 uS/cm (SS900)	100	0.0	100	0.0	20.8	1.70
SS900 Solvent Control	100	0.0	100	0.0	24.2	1.38
SS900 + 312.5 pptr Permethrin	100	0.0	100	0.0	21.6	1.28
SS900 + 625 pptr Permethrin	100	0.0	100	0.0	20.2	1.26
SS900 + 1250 pptr Permethrin	100	0.0	90	10.0	16.3	1.89
SS900 + 2500 pptr Permethrin	60	16.3	60	16.3	2.5	0.95
SS900 + 5000 pptr Permethrin	0	0.0	0	0.0	0.0	0.00
Hatchery Water @ 900 uS/cm (HW900)	100	0.0	100	0.0	26.4	1.53
HW900 Solvent Control	100	0.0	100	0.0	27.9	2.00
HW900 + 312.5 pptr Permethrin	100	0.0	100	0.0	25.0	2.10
HW900 + 625 pptr Permethrin	100	0.0	100	0.0	23.8	1.91
HW900 + 1250 pptr Permethrin	100	0.0	90	10.0	16.3	2.22
HW900 + 2500 pptr Permethrin	60	16.3	60	16.3	2.1	1.05
HW900 + 5000 pptr Permethrin	0	0.0	0	0.0	0.0	0.00

1. Highlighted cells indicate statistically significant reductions in survival or reproduction compared to the solvent control. Data were analyzed using USEPA standard statistical protocols.

Table J 8.2. Nominal 7-day effect concentrations of permethrin in a *C. dubia* test initiated on 12/02/09.

Endpoint	Matrix	Permethrin (pptr)						
		NOEC	LOEC	LC10/EC10		LC50 (survival) / EC25 (reproduction)		PMSD
				Estimate	95% C.I.	Estimate	95% C.I.	
96-hour Survival	SSEPAMH @ 900 uS/cm	2500	5000	1487	1380 - 2500	2806	2051 - 3402	NA
	Hatchery Water @ 900 uS/cm	2500	5000	1487	1380 - 2500	2806	2051 - 3402	NA
7-day Survival	SSEPAMH @ 900 uS/cm	2500	5000	1250	787.5 - 2500	2806	1895 - 3402	NA
	Hatchery Water @ 900 uS/cm	2500	5000	1250	787.5 - 2500	2806	1895 - 3402	NA
7-day Reproduction	SSEPAMH @ 900 uS/cm	625	1250	209.6	12.25 - 725.7	899.8	498 - 1354	18.02%
	Hatchery Water @ 900 uS/cm	625	1250	251.1	9.7 - 771	815.3	357 - 1268	21.44%

Table J 8.3. Water chemistry during a *C. dubia* 7-day test initiated 12/02/09 evaluating the toxicity of permethrin in both conductivity-adjusted synthetic water and water from the UC Davis Fish Conservation and Culture Laboratory, Skinner Fish Protection Facility, Byron, California.

Treatment	Initial EC (uS/cm)	Temp (°C)		DO (mg/L)		pH		Hardness (mg/L as $CaCO_3$)	Alkalinity (mg/L as $CaCO_3$)
		Min	Max	Min	Max	Min	Max		
SSEPAMH	224	21.0	25.7	7.2	8.8	7.8	8.37	88	62
SSEPAMH @ 900 uS/cm (SS900)	848	21.7	25.7	7.4	8.7	7.71	8.27	180	58
SS900 Solvent Control	859	21.8	25.8	6.8	8.8	7.76	8.27	-	-
SS900 + 312.5 pptr Permethrin	890	21.5	25.7	6.8	8.7	7.77	8.25	-	-
SS900 + 625 pptr Permethrin	871	21.3	25.6	7.2	8.9	7.81	8.3	-	-
SS900 + 1250 pptr Permethrin	897	21.7	25.6	7.1	8.9	7.79	8.33	-	-
SS900 + 2500 pptr Permethrin	838	21.7	25.5	7.2	8.8	7.78	8.33	-	-
SS900 + 5000 pptr Permethrin	895	24.0	24.8	7.4	8.6	7.79	7.95	-	-
Hatchery Water @ 900 uS/cm (HW900)	840	21.8	25.5	7.2	9.7	7.83	8.57	148	72
HW900 Solvent Control	868	22.0	25.5	6.7	9.4	7.82	8.51	-	-
HW900 + 312.5 pptr Permethrin	834	21.7	25.4	6.8	9.3	7.85	8.51	-	-
HW900 + 625 pptr Permethrin	835	21.7	25.3	6.9	9.3	7.83	8.52	-	-
HW900 + 1250 pptr Permethrin	870	21.2	25.3	7.0	9.3	7.86	8.54	-	-
HW900 + 2500 pptr Permethrin	840	21.0	25.3	7.2	9.4	7.89	8.51	-	-
HW900 + 5000 pptr Permethrin	870	23.9	24.9	7.6	8.6	7.92	7.97	-	-

Table J 9.1. Results of a *C. dubia* 7-day test initiated 3/03/10 evaluating the toxicity of copper in conductivity-adjusted synthetic water and in water collected from the UC Davis Fish Conservation and Culture Laboratory, Skinner Fish Protection Facility, Byron, California..

Treatment	Dissolved Copper (mg/L)	96-hour Survival (%)[1]		7-day Survival (%)[1]		Reproduction (offspring)[1]	
		Mean	SE	Mean	SE	Mean	SE
SDEPAMH	-	100	0	100	0	26	0.71
SDEPAMH @ 900 µS/cm (SD900)	ND	100	0	100	0	25.1	1.32
SD900 + 37.5 ppb Copper	34.9	90	10	90	10	17.8	2.45
SD900 + 75 ppb Copper	72.6	30	15	10	10	1.4	0.91
SD900 + 150 ppb Copper	151	0	0	0	0	0	0.00
SD900 + 300 ppb Copper	305	0	0	0	0	0	0.00
SD900 + 600 ppb Copper	-	0	0	0	0	0	0.00
SD900 + 1200 ppb Copper	-	0	0	0	0	0	0.00
Hatchery Water @ 900 µS/cm (HW900)	-	100	0	100	0	23	1.20
HW900 + 37.5 ppb Copper	-	100	0	100	0	24.7	0.62
HW900 + 75 ppb Copper	-	100	0	100	0	19.7	2.54
HW900 + 150 ppb Copper	-	70	15	70	15	7.5	1.93
HW900 + 300 ppb Copper	-	0	0	0	0	0.0	0.00
HW900 + 600 ppb Copper	-	0	0	0	0	0.0	0.00
HW900 + 1200 ppb Copper	-	0	0	0	0	0.0	0.00

1. Highlighted cells indicate statistically significant reductions in survival or reproduction compared to the laboratory control. Data were analyzed using USEPA standard statistical protocols.

Table J 9.2. Nominal 7-day effect concentrations of copper in a *C. dubia* test initiated on 3/03/10.

Endpoint	Matrix	NOEC	LOEC	LC10/EC10		LC50 (survival) / EC25 (reproduction)		PMSD
				Estimate	95% C.I.	Estimate	95% C.I.	
96-hour Survival	SDEPAMH @ 900 uS/cm	37.5	75	37.5	2.4 - 43.1	59.6	49.5 - 84.2	NA
	Hatchery Water @ 900 uS/cm	150	300	94.5	84.2 - 150	182.9	133.7 - 204.2	NA
7-day Survival	SDEPAMH @ 900 uS/cm	37.5	75	37.5	2.4 - 40.9	53.1	47.3 – 59.5	NA
	Hatchery Water @ 900 uS/cm	150	300	106.1	86.2 – 160.8	194.6	150 – 212.2	NA
Reproduction	SDEPAMH @ 900 uS/cm	< 37.5	37.5	2.20	1.1 - 11.8	17.1	5.3 - 40.9	19.0%
	Hatchery Water @ 900 uS/cm	75	150	55.90	44.0 - 83.2	83.2	58.5 - 98.2	22.7%

Table J 9.3. Measured 7-day effect concentrations of copper in a *C. dubia* test initiated on 3/03/10.

Endpoint	Matrix	NOEC	LOEC	LC10/EC10		LC50 (survival) / EC25 (reproduction)		PMSD
				Estimate	95% C.I.	Estimate	95% C.I.	
96-hour Survival	SDEPAMH @ 900 uS/cm	34.9	72.6	34.9	2.3 - 40.4	56.9	44.6 - 82.1	NA
	Hatchery Water @ 900 uS/cm	151	305	92.7	82.1 - 162	184.6	133.7 - 214.7	NA
7-day Survival	SDEPAMH @ 900 uS/cm	34.9	72.6	34.9	2.3 - 38.8	50.4	43.1 - 59.0	NA
	Hatchery Water @ 900 uS/cm	151	305	92.7	82.1 - 162	184.6	133.7 - 214.7	NA
Reproduction	SDEPAMH @ 900 uS/cm	< 34.9	34.9	2.4	1.1 - 21.2	20.7	5.1 - 39.4	19.0%
	Hatchery Water @ 900 uS/cm	72.6	151	53.20	41.5 - 80.2	81.0	54.2 - 95.6	22.7%

Table J 9.4. Summary of water chemistry during a *C. dubia* toxicity test initiated on 3/03/10 evaluating the toxicity of copper in conductivity-adjusted synthetic water and in water collected from the UC Davis Fish Conservation and Culture Laboratory, Skinner Fish Protection Facility, Byron, California.

Treatment	Initial EC (uS/cm)	Temp (°C)		DO (mg/L)		pH		Hardness (mg/L as CaCO3)	Alkalinity (mg/L as CaCO3)
		Min	Max	Min	Max	Min	Max		
SDEPAMH	292	24.1	26.2	6.7	8.3	7.77	8.27	92	56
SDEPAMH @ 900 µS/cm (SD900)	892	23.7	26.2	7.2	8.6	7.74	8.11	156	60
SD900 + 37.5 ppb Copper	880	23.8	26.1	7.2	8.5	7.75	8.16	-	-
SD900 + 75 ppb Copper	885	23.9	26.4	7.3	8.5	7.69	8.13	-	-
SD900 + 150 ppb Copper	853	24.0	25.8	7.3	8.4	7.72	8.02	-	-
SD900 + 300 ppb Copper	883	23.7	25.2	7.7	8.4	7.74	7.97	-	-
SD900 + 600 ppb Copper	874	23.8	25.2	7.8	8.4	7.64	8.02	-	-
SD900 + 1200 ppb Copper	891	23.8	25.3	7.8	8.2	7.57	8.01	-	-
Hatchery Water @ 900 µS/cm (HW900)	868	24.0	26.0	7.2	8.5	7.59	8.28	128	88
HW900 + 37.5 ppb Copper	870	23.8	26.1	7.3	8.6	7.66	8.24	-	-
HW900 + 75 ppb Copper	879	23.7	26.3	7.1	8.5	7.66	8.25	-	-
HW900 + 150 ppb Copper	872	23.8	26.1	7.2	8.5	7.65	8.23	-	-
HW900 + 300 ppb Copper	872	23.7	25.6	7.3	8.3	7.62	8.23	-	-
HW900 + 600 ppb Copper	863	24.4	25.2	8.0	8.4	7.59	8.11	-	-
HW900 + 1200 ppb Copper	870	23.8	25.2	8.1	8.2	7.57	8.17	-	-

Table J 10.1. Results of a *C. dubia* 7-day test initiated 3/18/10 evaluating the toxicity of bifenthrin in both conductivity-adjusted synthetic water and water from the UC Davis Fish Conservation and Culture Laboratory, Skinner Fish Protection Facility, Byron, California.

Treatment	Measured Bifenthrin (pptr in Milli-Q)	96-hour Survival (%)[1]		7-day Survival (%)[1]		Reproduction (offspring)[1]	
		Mean	SE	Mean	SE	Mean	SE
SDEPAMH	-	100	0	100	0.0	6.8	1.60
SDEPAMH @ 900 uS/cm (SD900)	-	100	0	100	0.0	15.8	1.96
SD900 Solvent Control	ND	100	0	100	0.0	17.7	2.15
SD900 + 125 pptr Bifenthrin	68	100	0	100	0.0	17.8	2.89
SD900 + 250 pptr Bifenthrin	179	100	0	100	0.0	20.3	1.67
SD900 + 500 pptr Bifenthrin	288	100	0	90	10.0	11.3	1.93
SD900 + 1000 pptr Bifenthrin	432	70	15	0	0.0	0.0	0.00
SD900 + 2000 pptr Bifenthrin	752	0	0	0	0.0	0.0	0.00
SD900 + 4000 pptr Bifenthrin	-	0	0	0	0.0	0.0	0.00
SD900 + 8000 pptr Bifenthrin	-	0	0	0	0.0	0.0	0.00
Hatchery Water @ 900 (HW900)	-	100	0	100	0.0	24.7	1.21
HW900 Solvent Control	ND	100	0	100	0.0	16.1	1.25
HW900 + 250 pptr Bifenthrin	179	100	0	90	10.0	17.2	1.16
HW900 + 500 pptr Bifenthrin	288	100	0	40	16.3	9.0	1.15
HW900 + 1000 pptr Bifenthrin	432	40	16	0	0.0	0.0	0.00
HW900 + 2000 pptr Bifenthrin	752	0	0	0	0.0	0.0	0.00
HW900 + 4000 pptr Bifenthrin	-	0	0	0	0.0	0.0	0.00
HW900 + 8000 pptr Bifenthrin	-	0	0	0	0.0	0.0	0.00

Table J 10.2. Nominal 7-day effect concentrations of bifenthrin in a *C. dubia* test initiated on 3/18/10.

Endpoint	Matrix	Bifenthrin (pptr)						
		NOEC	LOEC	LC10/EC10		LC50 (survival) / EC25 (reproduction)		PMSD
				Estimate	95% C.I.	Estimate	95% C.I.	
96-h Survival	SDEPAMH @ 900 uS/cm	1000	2000	630	561.3 - 1072	1219	890.9 - 1414	NA
	Hatchery Water @ 900 uS/cm	500	1000	561.3	540 - 630	890.9	734.9 - 1219	NA
7-day Survival	SDEPAMH @ 900 uS/cm	500	1000	500.0	315 - 535.9	680.5	609.6-707.2	NA
	Hatchery Water @ 900 uS/cm	250	500	280.6	270-315	445.5	367.5-609.6	NA
7-day Reproduction	SDEPAMH @ 900 uS/cm	500	1000	298.4	15.2 - 344.4	388.9	298.2 - 520.4	37.9%
	Hatchery Water @ 900 uS/cm	250	500	290.7	78.1 - 315.3	364.6	310.7 - 446.6	20.9%

Table J 10.3. Measured 7-day effect concentrations of bifenthrin in a *C. dubia* test initiated on 3/18/10.

Endpoint	Matrix	Bifenthrin (pptr)						
		NOEC	LOEC	LC10/EC10		LC50 (survival) / EC25 (reproduction)		PMSD
				Estimate	95% C.I.	Estimate	95% C.I.	
96-h Survival	SDEPAMH @ 900 uS/cm	432	752	329.7	308.1 - 432	506.2	403.8 - 552.7	NA
	Hatchery Water @ 900 uS/cm	288	432	308.1	301.3 - 329.7	403.8	360.8 - 506.2	NA
7-day Survival	SDEPAMH @ 900 uS/cm	288	432	288.0	209.8 - 299.9	344.9	323.4 - 352.7	NA
	Hatchery Water @ 900 uS/cm	179	288	193.8	188.7 - 209.8	266.1	233.2 - 323.4	NA
7-day Reproduction	SDEPAMH @ 900 uS/cm	288	432	203.1	8.0 – 240.0	245.4	203 – 302	37.9%
	Hatchery Water @ 900 uS/cm	179	288	198.5	86.9 - 208.7	231.9	211.6 - 262.9	20.9%

Table J 10.4. Water chemistry during a *C. dubia* 7-day test initiated 3/18/10 evaluating the toxicity of bifenthrin in both conductivity-adjusted synthetic water and water from the UC Davis Fish Conservation and Culture Laboratory, Skinner Fish Protection Facility, Byron, California.

Treatment	Initial EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH	Hardness (mg/L as $CaCO_3$)	Alkalinity (mg/L as $CaCO_3$)
SDEPAMH	284	23.7	26.7	6.6	7.9	7.77	8.34	84	60
SDEPAMH @ 900 uS/cm (SD900)	843	15.1	26.3	6.8	8.1	7.79	8.14	126	57
SD900 Solvent Control	850	23.2	26.3	5.7	8.2	7.58	8.14	-	-
SD900 + 125 pptr Bifenthrin	828	23.2	26.4	5.7	8.2	7.62	8.20	-	-
SD900 + 250 pptr Bifenthrin	832	23.3	26.5	5.9	8.3	7.59	8.18	-	-
SD900 + 500 pptr Bifenthrin	837	23.0	29.3	5.2	8.3	7.63	8.15	-	-
SD900 + 1000 pptr Bifenthrin	827	23.2	27.1	5.3	8.2	7.61	8.19	-	-
SD900 + 2000 pptr Bifenthrin	840	23.1	26.4	6.3	8.2	7.61	8.20	-	-
SD900 + 4000 pptr Bifenthrin	831	23.2	26.5	6.7	8.2	7.82	8.08	-	-
SD900 + 8000 pptr Bifenthrin	838	23.9	26.4	6.9	7.8	7.81	8.05	-	-
Hatchery Water @ 900 (HW900)	831	23.3	27.0	6.7	8.4	7.89	8.26	-	-
HW900 Solvent Control	893	23.3	26.9	3.2	8.4	7.54	8.26	-	-
HW900 + 250 pptr Bifenthrin	841	23.3	26.9	3.1	8.5	7.52	8.26	-	-
HW900 + 500 pptr Bifenthrin	852	23.2	27.0	3.3	9.6	7.55	8.26	-	-
HW900 + 1000 pptr Bifenthrin	836	23.2	26.9	3.1	8.5	7.59	8.26	-	-
HW900 + 2000 pptr Bifenthrin	862	23.3	26.5	4.4	8.5	7.65	8.22	-	-
HW900 + 4000 pptr Bifenthrin	842	23.2	26.2	4.2	8.5	7.66	8.23	-	-
HW900 + 8000 pptr Bifenthrin	852	24.1	25.9	5.4	8.0	7.73	8.19	-	-

Table J 11.1. Results of a *C. dubia* 7-day test initiated 4/07/10 evaluating the toxicity of cyfluthrin in both conductivity-adjusted synthetic water and water from the UC Davis Fish Conservation and Culture Laboratory, Skinner Fish Protection Facility, Byron, California.

Treatment	Measured Cyfluthrin (pptr in Milli-Q)	96-hour Survival (%)[1]		7-day Survival (%)[1]		Reproduction (offspring)[1]	
		Mean	SE	Mean	SE	Mean	SE
SDEPAMH	-	100	0	100	0	18.9	1.5
SDEPAMH @ 900 uS/cm (SD900)	-	80	13	80	13	11.4	3.1
SDEPAMH Solvent Control	ND	90	10	90	10	14.3	2.7
SD900 + 62.5 pptr Cyfluthrin	35	100	0	100	0	14.6	2.5
SD900 + 125 pptr Cyfluthrin	108	100	0	100	0	11.8	1.9
SD900 + 250 pptr Cyfluthrin	268	90	10	90	10	9.7	1.6
SD900 + 500 pptr Cyfluthrin	515	100	0	90	10	2.0	0.9
SD900 + 1000 pptr Cyfluthrin	985	0	0	0	0	0.0	0.0
SD900 + 2000 pptr Cyfluthrin	-	0	0	0	0	0.0	0.0
SD900 + 4000 pptr Cyfluthrin	-	0	0	0	0	0.0	0.0
Hatchery Water @ 900 uS/cm (HW900)	-	100	0	100	0	27.5	1.3
HW900 Solvent Control	-	100	0	100	0	27.3	1.4
HW900 + 125 pptr Cyfluthrin	108	100	0	100	0	21.9	1.3
HW900 + 250 pptr Cyfluthrin	268	100	0	100	0	14.1	1.6
HW900 + 500 pptr Cyfluthrin	515	100	0	100	0	3.2	0.8
HW900 + 1000 pptr Cyfluthrin	985	0	0	0	0	0.0	0.0
HW900 + 2000 pptr Cyfluthrin	-	0	0	0	0	0.0	0.0
HW900 + 4000 pptr Cyfluthrin	-	0	0	0	0	0.0	0.0

Table J 11.2. Nominal 7-day effect concentrations of cyfluthrin in a *C. dubia* test initiated on 4/07/10.

Endpoint	Matrix	Cyfluthrin (pptr)						
		NOEC	LOEC	LC10/EC10		LC50 (survival) / EC25 (reproduction)		PMSD
				Estimate	95% C.I.	Estimate	95% C.I.	
96-hour Survival	SDEPAMH @ 900 uS/cm	500	1000	530.1	222 - 535.9	702.9	674.3 - 707.2	NA
	Hatchery Water @ 900 uS/cm	500	1000	535.9	-	707.2	-	NA
7-day Survival	SDEPAMH @ 900 uS/cm	500	1000	511.7	177 - 536	689.2	610 - 707	NA
	Hatchery Water @ 900 uS/cm	500	1000	535.9	-	707.2	-	NA
7-day Reproduction	SDEPAMH @ 900 uS/cm	250	500	91.3	3 - 203	171.8	25 - 282	45.0%
	Hatchery Water @ 900 uS/cm	< 125	125	9.8	4 - 96	140.0	49 - 177	14.5%

Table J 11.3. Measured 7-day effect concentrations of cyfluthrin in a *C. dubia* test initiated on 4/07/10.

Endpoint	Matrix	Cyfluthrin (pptr)						
		NOEC	LOEC	LC10/EC10		LC50 (survival) / EC25 (reproduction)		PMSD
				Estimate	95% C.I.	Estimate	95% C.I.	
96-hour Survival	SDEPAMH @ 900 uS/cm	515	985	543.9	229.4 - 549.5	708.2	681.3 - 712.3	NA
	Hatchery Water @ 900 uS/cm	515	985	549.5	-	712.3	-	NA
7-day Survival	SDEPAMH @ 900 uS/cm	515	985	537.8	183 - 550	703.8	620 -712	NA
	Hatchery Water @ 900 uS/cm	515	985	549.5	-	712.3	-	NA
7-day Reproduction	SDEPAMH @ 900 uS/cm	268	515	64.9	1.8 - 270.9	163.9	12.0 - 307.7	45.0%
	Hatchery Water @ 900 uS/cm	<108	108	9.7	3.6 - 112.5	127.6	43.3 - 183.3	14.5%

Table J 11.4. Water chemistry during a *C. dubia* 7-day test initiated 4/07/10 evaluating the toxicity of cyfluthrin in both conductivity-adjusted synthetic water and water from the UC Davis Fish Conservation and Culture Laboratory, Skinner Fish Protection Facility, Byron, California.

Treatment	Initial EC (uS/cm)	Min Temp (°C)	Max Temp (°C)	Min DO (mg/L)	Max DO (mg/L)	Min pH	Max pH	Hardness (mg/L as $CaCO_3$)	Alkalinity (mg/L as $CaCO_3$)
SDEPAMH	292	24.0	26.4	6.8	8.1	7.85	8.51	92	56
SDEPAMH @ 900 uS/cm (SD900)	871	23.5	26.4	6.6	8.3	7.80	8.47	88	56
SDEPAMH Solvent Control	871	23.7	26.5	6.2	8.3	7.78	8.43	-	-
SD900 + 62.5 pptr Cyfluthrin	870	23.5	26.6	6.1	8.3	7.78	8.45	-	-
SD900 + 125 pptr Cyfluthrin	872	23.6	26.6	5.9	8.3	7.74	8.43	-	-
SD900 + 250 pptr Cyfluthrin	868	23.7	26.6	6.1	8.2	7.74	8.48	-	-
SD900 + 500 pptr Cyfluthrin	874	23.6	26.6	6.2	8.3	7.74	8.51	-	-
SD900 + 1000 pptr Cyfluthrin	873	23.7	26.7	7.0	8.3	7.81	8.52	-	-
SD900 + 2000 pptr Cyfluthrin	874	23.7	27.0	6.9	8.4	7.83	8.50	-	-
SD900 + 4000 pptr Cyfluthrin	874	23.7	27.2	6.5	8.2	7.84	8.44	-	-
Hatchery Water @ 900 uS/cm (HW900)	878	23.9	26.7	6.4	8.6	7.80	8.37	132	92
HW900 Solvent Control	878	23.9	26.6	4.0	8.2	7.58	8.39	-	-
HW900 + 125 pptr Cyfluthrin	881	23.8	26.7	3.8	8.0	7.57	8.39	-	-
HW900 + 250 pptr Cyfluthrin	877	23.8	26.6	4.0	8.1	7.62	8.37	-	-
HW900 + 500 pptr Cyfluthrin	883	23.8	26.8	4.1	8.6	7.64	8.39	-	-
HW900 + 1000 pptr Cyfluthrin	879	23.8	26.9	5.7	8.1	7.82	8.41	-	-
HW900 + 2000 pptr Cyfluthrin	884	24.3	26.8	6.4	8.2	7.82	8.30	-	-
HW900 + 4000 pptr Cyfluthrin	891	23.9	26.9	6.3	8.2	7.81	8.40	-	-

Appendix K

Quality Assurance/Quality Control: Relative Percent Differences

Table K1. Individual Relative Percent Differences of water chemistry measurements between Site 609 and its duplicate

Collection Date	Analyte					
	EC	DO	pH	Hardness	Alkalinity	Ammonia
May 7, 2008	2.43 2.25	1.24 2.74 2.38 1.38 2.63 0.00 2.38 0.00 1.29 1.16	2.12 0.00 0.63 0.00 1.78 0.38	8.45	1.83	11.76
June 3, 2008	0.53 0.88	0.00 0.00 3.77 0.00 2.56 7.14 7.50 5.19 5.33 2.41	0.88 0.38 0.13 0.12 0.38 0.12	4.65	3.64	15.38
August 27, 2008	3.70 2.14	3.73 4.38 0.00 1.29 2.78 3.68 3.55 2.67 0.00 0.00	1.77 0.13 0.51 1.16 0.25 0.65	9.76	0.00	66.67
September 10, 2008	0.42 0.71	0.00 1.40 2.47 2.78 2.70 1.26 0.00 2.53 4.14 0.00	0.00 0.00 0.26 0.25 0.90 0.64	2.35	1.80	0.00
December 17, 2008	7.53 1.08	1.29 0.00 5.00 0.00 1.29 10.13 3.82 3.87 3.87 3.73	1.51 0.51 0.25 0.51 0.63 0.50	5.13	1.07	0.00
May 27, 2009	0.81 1.60	1.24 1.31 2.41 2.67 1.29 5.78 0.00 2.60 1.32 3.55	0.50 0.38 0.38 0.25 0.64 0.38	0.00	9.09	6.45
October 28, 2009	1.98 1.99	2.50 0.00 1.31 9.76 6.37 2.30 0.00 1.34 0.00 10.00	0.51 0.00 0.25 0.77 0.13 0.64	2.47	4.55	8.33

Table K2. Average Relative Percent Differences of water chemistry measurements between Site 609 and its duplicate

Collection Date	EC			DO			pH		
	Sample Size	Average	SD	Sample Size	Average	SD	Sample Size	Average	SD
May 7, 2008	2	2.34	0.12	10	1.52	1.01	6	0.82	0.92
June 3, 2008	2	0.70	0.25	10	3.39	2.87	6	0.34	0.30
August 27, 2008	2	2.92	1.10	10	2.21	1.73	6	0.74	0.62
September 10, 2008	2	0.56	0.20	10	1.73	1.43	6	0.34	0.36
December 17, 2008	2	4.30	4.56	10	3.30	2.99	6	0.65	0.44
May 27, 2009	2	1.20	0.56	10	2.22	1.61	6	0.42	0.13
October 28, 2009	2	1.99	0.01	10	3.36	3.92	6	0.38	0.30

Table K3. Individual Relative Percent Differences of water chemistry measurements between Site Hood and its duplicate

Collection Date	Analyte					
	EC	DO	pH	Hardness	Alkalinity	Ammonia
March 12, 2008	0.38	6.06	3.92	4.88	2.41	190.00
	1.16	2.44	0.00			
		0.00	1.13			
		2.44	0.50			
		0.00	2.53			
		2.35	0.90			
		2.63				
		0.00				
		1.23				
		1.29				
September 23, 2008	3.13	0.00	0.64	0.00	0.00	7.14
	2.93	3.55	2.55			
		4.03	2.72			
		1.20	0.26			
		5.48	1.06			
		0.00	0.91			
		2.82				
		2.41				
		2.82				
		2.67				
September 3, 2009	0.18	1.23	1.38	0.00	2.82	200.00
	7.84	1.24	1.17			
		1.31	0.49			
		1.17	0.38			
		4.08	2.94			
		2.35	0.13			
		2.70				
		3.51				
		0.00				
		8.00				
September 15, 2009	0.47	0.00	0.25	3.77	0.00	200.00
	0.94	2.41	1.06			
		6.71	0.76			
		2.44	1.79			
		5.41	2.99			
		1.29	0.39			
		2.56				
		3.51				
		2.63				
		0.00				

Table K4. Average Relative Percent Differences of water chemistry measurements between Site Hood and its duplicate

Collection Date	EC			DO			pH		
	Sample Size	Average	SD	Sample Size	Average	SD	Sample Size	Average	SD
March 12, 2008	2	0.69	0.45	10	1.84	1.83	6	1.50	1.46
September 23, 2008	2	3.03	0.15	10	2.50	1.72	6	1.36	1.03
September 3, 2009	2	4.01	5.42	10	2.56	2.27	6	1.08	1.03
September 15, 2009	2	0.71	0.34	10	2.70	2.13	6	1.21	1.03

Table K5. Individual Relative Percent Differences of water chemistry measurements between Site 711 and its duplicate

Collection Date	Analyte					
	EC	DO	pH	Hardness	Alkalinity	Ammonia
February 29, 2008	11.53 4.15	7.32 5.78 3.92 4.76 2.56 2.44 0.00 2.30 1.31 1.34	1.80 1.16 0.26 0.63 0.77 0.39	6.90	3.51	2.67
October 23, 2008	0.05 1.36	1.21 0.00 0.00 2.30 2.53 3.51 0.00 1.20 2.78 0.00	1.13 0.26 0.13 0.63 0.40 0.00	0.00	2.99	2.99
January 22, 2009	1.80 13.26	5.85 3.64 0.00 2.41 2.60 3.43 5.06 2.67 1.48 1.32	2.64 0.25 0.49 1.01 0.63 0.62	0.00	8.28	14.29

Table K6. Average Relative Percent Differences of water chemistry measurements between Site 711 and its duplicate

Collection Date	EC			DO			pH		
	Sample Size	Average	SD	Sample Size	Average	SD	Sample Size	Average	SD
February 29, 2008	2	7.84	5.22	10	3.17	2.25	6	0.83	0.57
October 23, 2008	2	0.71	0.93	10	1.35	1.35	6	0.42	0.41
January 22, 2009	2	7.53	8.11	10	2.85	1.75	6	0.94	0.87

Table K7. Individual Relative Percent Differences of water chemistry measurements between Site 508 and its duplicate

Collection Date	Analyte					
	EC	DO	pH	Hardness	Alkalinity	Ammonia
February 12, 2008	8.84 9.02	1.16 2.35 2.56 1.26 1.36 1.16 5.26 7.23	1.00 1.01 1.36 1.16 5.26 7.23	4.08	2.53	0.00
August 13, 2008	2.59 4.30	1.26 1.24 1.36 0.00 1.32 2.47 2.82 3.73 6.90 4.20	0.51 0.26 0.00 1.03 0.51 1.31	0.00	2.90	66.67
June 24, 2009	6.23 5.46	1.26 3.59 2.82 2.41 0.00 5.26 2.44 3.51 22.56 2.70	1.53 1.39 0.41 2.51 2.80 1.60	0.00	0.00	22.22

Table K8. Average Relative Percent Differences of water chemistry measurements between Site 508 and its duplicate

Collection Date	EC			DO			pH		
	Sample Size	Average	SD	Sample Size	Average	SD	Sample Size	Average	SD
February 12, 2008	2	8.93	0.12	8	2.48	2.18	6	0.56	0.39
August 13, 2008	2	3.45	1.21	10	2.53	2.00	6	0.60	0.49
June 24, 2009	2	5.85	0.54	10	4.65	6.44	6	1.71	0.86

Table K9. Individual Relative Percent Differences of water chemistry measurements between Site 340 and its duplicate

Collection Date	Analyte					
	EC	DO	pH	Hardness	Alkalinity	Ammonia
January 30, 2008	2.21 0.94	0.00 3.72 5.00 3.55 1.21 2.47 1.24 1.27 1.32 1.29	0.13 0.00 0.25 0.13 0.13 0.13	12.12	2.47	3.92
March 4, 2009	1.47 4.27	4.82 1.26 5.41 1.26 1.26 7.06 1.27 3.43 6.54 2.53	0.38 0.13 0.13 0.00 0.00 0.00	10.91	2.15	0.00

Table K10. Average Relative Percent Differences of water chemistry measurements between Site 340 and its duplicate

Collection Date	EC			DO			pH		
	Sample Size	Average	SD	Sample Size	Average	SD	Sample Size	Average	SD
January 30, 2008	2	1.58	0.90	10	2.11	1.53	6	0.13	0.08
March 4, 2009	2	2.87	1.98	10	3.48	2.31	6	0.11	0.15

Table K11. Individual Relative Percent Differences of water chemistry measurements between Site Rough & Ready and its duplicate

Collection Date	Analyte					
	EC	DO	pH	Hardness	Alkalinity	Ammonia
June 17, 2008	3.98	1.29	1.74	5.88	2.35	9.52
	2.27	2.41	0.89			
		5.33	1.52			
		2.38	0.38			
		2.78	0.38			
		0.61	0.75			
		2.56				
		2.41				
		1.36				
		2.27				
September 9, 2008	2.03	3.77	0.51	2.60	0.00	66.67
	2.18	1.21	0.00			
		2.63	1.25			
		2.44	1.24			
		0.00	0.63			
		0.00	0.49			
		1.29				
		2.47				
		1.36				
		1.34				
December 2, 2008	5.56	1.27	3.22	6.74	1.77	0.00
	5.68	1.20	0.12			
		2.50	0.61			
		3.68	0.12			
		3.77	0.12			
		5.99	1.10			
		17.14				
		3.68				
		0.00				
		0.00				
February 17, 2009	0.00	3.77	1.33	3.70	66.67	151.02
	1.95	0.00	0.86			
		5.33	0.12			
		0.00	0.37			
		3.92	0.50			
		7.69	0.00			
		1.26				
		0.00				
		5.13				
		2.70				

Table K12. Average Relative Percent Differences of water chemistry measurements of Site Rough & Ready and its duplicate

Collection Date	EC			DO			pH		
	Sample Size	Average	SD	Sample Size	Average	SD	Sample Size	Average	SD
June 17, 2008	2	3.13	1.21	10	2.11	1.46	6	0.95	0.56
September 9, 2008	2	5.62	0.06	10	3.92	5.02	6	0.88	1.21
December 2, 2008	2	2.11	0.10	10	1.65	1.19	6	0.50	0.44
February 17, 2009	2	0.97	1.38	10	2.98	2.66	6	0.53	0.50

Table K13. Individual Relative Percent Differences of water chemistry measurements between Site 602 and its duplicate

Collection Date	Analyte					
	EC	DO	pH	Hardness	Alkalinity	Ammonia
July 30, 2008	2.42	1.31	0.38	0.00	2.20	13.33
	0.81	1.27	0.00			
		2.78	0.00			
		2.50	0.00			
		5.41	0.00			
		2.47	0.00			
		2.78				
		0.00				
		4.14				
		0.00				
January 7, 2009	3.94	2.41	0.26	4.48	0.00	4.08
	2.96	0.00	0.13			
		1.31	0.51			
		1.21	0.00			
		6.71	0.13			
		6.45	0.13			
		4.37				
		4.94				
		0.00				
		1.24				

Table K14. Average Relative Percent Differences of water chemistry measurements between Site 602 and its duplicate

Collection Date	EC			DO			pH		
	Sample Size	Average	SD	Sample Size	Average	SD	Sample Size	Average	SD
July 30, 2008	2	1.62	1.14	10	2.26	1.70	6	0.06	0.15
January 7, 2009	2	3.45	0.70	10	2.87	2.55	6	0.19	0.17

Table K15. Individual Relative Percent Differences of water chemistry measurements between Site 815 and its duplicate

Collection Date	Analyte					
	EC	DO	pH	Hardness	Alkalinity	Ammonia
July 17, 2008	1.58	2.35	1.02	6.45	6.25	18.18
	1.01	2.33	0.00			
		3.73	0.63			
		0.00	0.51			
		6.06	2.02			
		1.29	0.00			
		2.78				
		4.71				
		5.41				
		2.74				
June 11, 2009	0.62	2.44	2.25	6.45	3.51	0.00
	4.02	1.24	1.01			
		2.63	0.26			
		1.21	0.00			
		1.29	0.91			
		3.51	0.78			
		0.00				
		1.20				
		2.74				
		0.00				

Table K16. Average Relative Percent Differences of water chemistry measurements between Site 815 and its duplicate

Collection Date	EC			DO			pH		
	Sample Size	Average	SD	Sample Size	Average	SD	Sample Size	Average	SD
July 17, 2008	2	1.29	0.40	10	3.14	1.87	6	0.70	0.76
June 11, 2009	2	2.32	2.40	10	1.63	1.17	6	0.87	0.78

Table K17. Individual Relative Percent Differences of water chemistry measurements between Site 902 and its duplicate

Collection Date	Analyte					
	EC	DO	pH	Hardness	Alkalinity	Ammonia
July 3, 2008	0.14	4.82	1.51	5.41	5.88	22.22
	9.02	1.18	0.89			
		2.94	0.63			
		0.00	0.90			
		0.00	1.05			
		1.18	0.00			
		8.11				
		1.21				
		0.00				
		1.34				
April 23, 2009	1.19	0.00	0.00	4.88	0.00	100.00
	0.29	1.21	0.38			
		0.00	0.12			
		1.20	0.12			
		1.24	0.64			
		4.65	0.63			
		1.26				
		3.73				
		2.56				
		2.70				

Table K18. Average Relative Percent Differences of water chemistry measurements between Site 902 and its duplicate

Collection Date	EC			DO			pH		
	Sample Size	Average	SD	Sample Size	Average	SD	Sample Size	Average	SD
July 3, 2008	2	4.58	6.28	10	2.08	2.59	6	0.83	0.50
April 23, 2009	2	0.74	0.63	10	1.86	1.53	6	0.32	0.28

Table K19. Individual Relative Percent Differences of water chemistry measurements between Site 915 and its duplicate

Collection Date	Analyte					
	EC	DO	pH	Hardness	Alkalinity	Ammonia
September 25, 2008	0.44 1.46	0.00 1.16 0.00 1.26 1.14 4.03 0.87 1.32 2.56	0.62 0.00 0.50 0.13 0.26 0.38	0.00	2.47	100.00
August 6, 2009	12.80 32.30	2.44 1.21 2.74 3.73 1.32 0.00 2.74 2.38 1.27 2.78	0.62 0.66 0.52 0.27 0.39 0.39	8.70	3.77	0.00

Table K20. Average Relative Percent Differences of water chemistry measurements between Site 915 and its duplicate

Collection Date	EC			DO			pH		
	Sample Size	Average	SD	Sample Size	Average	SD	Sample Size	Average	SD
September 25, 2008	2	0.95	0.72	9	1.37	1.26	6	0.31	0.23
August 6, 2009	2	22.55	13.79	10	2.06	1.08	6	0.47	0.15

Table K21. Individual Relative Percent Differences of water chemistry measurements between Site Cache-Ulatis and its duplicate

Collection Date	Analyte					
	EC	DO	pH	Hardness	Alkalinity	Ammonia
November 6, 2008	0.86	2.47	1.51	8.45	0.00	33.33
	1.37	0.00	0.22			
		0.00	0.61			
		1.23	0.12			
		0.00	0.00			
		1.20	1.00			
		2.63				
		1.21				
		4.20				
		0.00				
October 15, 2009	6.90	0.00	1.18	0.00	0.00	14.63
	1.12	1.26	0.26			
		0.00	0.50			
		3.87	0.25			
		0.00	1.02			
		3.82	1.14			
		0.00				
		0.00				
		1.34				
		1.34				

Table K22. Average Relative Percent Differences of water chemistry measurements between Site Cache-Ulatis and its duplicate

Collection Date	EC			DO			pH		
	Sample Size	Average	SD	Sample Size	Average	SD	Sample Size	Average	SD
November 6, 2008	2	1.11	0.36	10	1.29	1.43	6	0.58	0.58
October 15, 2009	2	4.01	4.09	10	1.16	1.54	6	0.72	0.44

Table K23. Individual Relative Percent Differences of water chemistry measurements between Site Napa and its duplicate

Collection Date	Analyte					
	EC	DO	pH	Hardness	Alkalinity	Ammonia
November 4, 2008	1.73	1.29	0.00	3.17	2.20	5.41
	0.41	2.47	0.13			
		3.97	0.64			
		0.00	9.73			
		1.44	0.26			
		2.53	0.13			
		4.32				
		0.00				
		1.44				
		5.63				
July 7, 2009	1.43	4.65	0.79	3.63	0.00	120.00
	0.00	5.26	0.26			
		1.36	0.13			
		8.48	1.02			
		1.40	0.90			
		2.63	0.13			
		6.25				
		5.06				
		5.80				
		4.65				
October 29, 2009	0.41	2.30	1.30	1.55	3.77	4.65
	2.97	1.24	0.51			
		6.54	0.39			
		3.82	0.13			
		2.78	0.00			
		2.56	0.13			
		1.40				
		0.00				
		1.40				
		1.40				

Table K24. Average Relative Percent Differences of water chemistry measurements between Site Napa and its duplicate

Collection Date	EC			DO			pH		
	Sample Size	Average	SD	Sample Size	Average	SD	Sample Size	Average	SD
November 4, 2008	2	1.07	0.94	10	2.31	1.86	6	1.81	3.88
July 7, 2009	2	0.72	1.01	10	4.56	2.22	6	0.54	0.41
October 29, 2009	2	1.69	1.82	10	2.34	1.81	6	0.41	0.47

Table K25. Individual Relative Percent Differences of water chemistry measurements between Site Light 55 and its duplicate

Collection Date	Analyte					
	EC	DO	pH	Hardness	Alkalinity	Ammonia
October 9, 2008	3.79	0.00	0.25	4.88	2.35	28.57
	4.63	9.52	0.51			
		0.00	0.49			
		0.00	0.26			
		0.00	0.51			
		1.17	0.51			
		0.00				
		1.17				
		2.63				
		4.14				
February 4, 2009	0.30	2.53	1.61	8.00	7.41	14.08
	4.60	4.82	0.74			
		4.08	0.24			
		4.76	0.62			
		3.77	0.12			
		2.44	0.00			
		2.63				
		2.44				
		0.00				
		1.29				
August 6, 2009	0.04	3.64	0.12	19.05	2.90	185.20
	0.19	2.47	0.13			
		2.74	0.38			
		2.50	0.26			
		2.67	0.39			
		0.00	0.38			
		5.71				
		3.64				
		3.97				
		1.42				

Table K26. Average Relative Percent Differences of water chemistry measurements between Site Light 55 and its duplicate

Collection Date	EC			DO			pH		
	Sample Size	Average	SD	Sample Size	Average	SD	Sample Size	Average	SD
October 9, 2008	2	4.21	0.59	10	1.86	3.03	6	0.42	0.13
February 4, 2009	2	2.45	3.04	10	2.88	1.53	6	0.55	0.59
August 6, 2009	2	0.11	0.10	10	2.78	1.53	6	0.28	0.13

Table K27. Individual Relative Percent Differences of water chemistry measurements between Site Cache-Lindsey and its duplicate

Collection Date	Analyte					
	EC	DO	pH	Hardness	Alkalinity	Ammonia
October 9, 2008	0.27	1.24	0.13	6.06	0.00	0.00
	2.26	7.41	0.00			
		3.82	0.49			
		3.68	0.52			
		1.23	0.65			
		4.65	0.26			
		2.63				
		1.18				
		1.32				
		0.00				
July 9, 2009	0.13	1.18	0.00	19.35	0.00	5.13
	1.88	3.47	1.31			
		2.63	0.13			
		2.47	0.39			
		1.31	0.26			
		2.53	0.13			
		1.34				
		1.24				
		1.38				
		0.00				

Table K28. Average Relative Percent Differences of water chemistry measurements between Site Cache-Lindsey and its duplicate

Collection Date	EC			DO			pH		
	Sample Size	Average	SD	Sample Size	Average	SD	Sample Size	Average	SD
October 9, 2008	2	1.26	1.41	10	2.72	2.21	6	0.34	0.25
July 9, 2009	2	1.00	1.23	10	1.76	1.00	6	0.37	0.48

Table K29. Individual Relative Percent Differences of water chemistry measurements between Site Suisun and its duplicate

Collection Date	Analyte					
	EC	DO	pH	Hardness	Alkalinity	Ammonia
October 21, 2008	0.07	0.00	0.00	4.87	1.24	44.44
	0.43	0.00	0.63			
		0.00	0.63			
		3.55	0.75			
		3.97	0.00			
		0.00	0.62			
		0.00				
		0.00				
		1.34				
		1.34				
May 26, 2009	0.72	1.26	1.15	7.41	8.11	52.63
	0.94	1.24	0.12			
		0.00	0.50			
		1.21	0.62			
		2.33	0.37			
		2.60	0.12			
		2.38				
		1.32				
		2.63				

Table K30. Average Relative Percent Differences of water chemistry measurements between Site Suisun and its duplicate

Collection Date	EC			DO			pH		
	Sample Size	Average	SD	Sample Size	Average	SD	Sample Size	Average	SD
October 21, 2008	2	0.25	0.25	10	1.02	1.55	6	0.44	0.34
May 26, 2009	2	0.83	0.55	10	1.89	1.09	6	0.48	0.38

Table K31. Individual Relative Percent Differences of water chemistry measurements between Site 405 and its duplicate

Collection Date	Analyte					
	EC	DO	pH	Hardness	Alkalinity	Ammonia
June 24, 2009	1.78	5.13	0.39	0.00	2.35	29.79
	2.84	4.88	2.94			
		5.88	0.82			
		3.77	1.72			
		6.71	0.66			
		10.13	1.21			
		9.66				
		7.59				
		2.86				
		1.40				
November 10, 2009	0.68	4.71	0.26	18.46	4.26	22.22
	2.83	5.26	0.38			
		0.00	0.38			
		1.31	0.51			
		1.32	1.03			
		0.00	0.39			
		1.40				
		5.06				
		2.67				
		5.33				

Table K32. Average Relative Percent Differences of water chemistry measurements between Site 405 and its duplicate

Collection Date	EC			DO			pH		
	Sample Size	Average	SD	Sample Size	Average	SD	Sample Size	Average	SD
June 24, 2009	2	2.31	0.75	10	8.50	2.81	6	1.29	0.93
November 10, 2009	2	1.75	1.53	10	2.71	2.19	6	0.49	0.28