The Effects of Wetland Restoration on Mercury Bioaccumulation in the South Bay Salt Pond Restoration Project: Using the Biosentinel Toolbox to Monitor Changes Across Multiple Habitats and Spatial Scales

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ABSTRACT:
The South Bay Salt Pond Restoration Project's plans to convert salt ponds into tidal marsh habitat may result in changes to the distribution, availability, and bioaccumulation of methylmercury (MeHg) within the region, which is known to already have MeHg levels that exceed wildlife toxicity thresholds. Implementation of a robust monitoring network will allow restoration managers to document these changes simultaneously across multiple habitats, and may guide actions that compensate for unintended outcomes. The planned construction of a tidal breach in Pond A8 provides a unique opportunity to examine this approach. This proposal links benthic MeHg production potential, changes in water and sediment column Hg concentration and speciation with MeHg bioaccumulation in four key biosentinel species. Each biosentinel represents an important component of the local food web in the habitat mosaic, and each will provide direct evidence of Hg bioaccumulation across the landscape. A particular strength of this proposal is the collaboration of researchers involved, all of whom have been leading extensive mercury projects in the South Bay, and their ability to leverage baseline data to clearly demonstrate any changes in MeHg exposure as a result of the A8 management action. This study will provide critical information to ecosystem managers in guiding their future decisions regarding former salt pond management and wetland restoration activities.

BACKGROUND AND JUSTIFICATION:

A. Background: Work to Date

Two of the most significant anthropogenic changes in the San Francisco Bay (SFB) Estuary over the past 150 years are the loss of over 85% of fringing tidal wetlands (Goals Project 1999) and the contamination of the estuarine food web with mercury (Hg) (Greenfield *et al*. 2005). These impacts are particularly pronounced in the South Bay, which was historically fringed with extensive tidal marshes and which receives drainage from New Almaden, the largest historic Hg mining area in North America. Extensive restoration in the South Bay region aims to return much of the important ecosystem function these wetlands provided. However, high rates of methylmercury (MeHg; the most toxic form of Hg) production, export, and bioaccumulation have been associated with wetlands relative to other water bodies (Hurley *et al.* 1995, Krabbenhoft *et al.* 1999, Waldron *et al.* 2000, Yee *et al.* 2008). Thus, the potential exists to increase Hg bioavailability in the South Bay as former salt ponds are restored to tidal marsh. This is a particularly important concern, because Hg concentrations in tissues and eggs of waterbirds in the South Bay currently exceed toxicological thresholds (Figure 1; Eagles-Smith *et al.* 2009, Eagles-Smith and Ackerman 2008), and there is evidence that Hg may be impairing egg hatchability and chick survival (Figure 2; Ackerman and Eagles-Smith 2008, Ackerman *et al.* 2008a). Thus, any increase in MeHg production and subsequent bioaccumulation in waterbirds may have a substantial impact to their reproduction.

One of the first major changes in the restoration process is the planned breach of Pond A8, to return it to muted tidal action. This breach will be in the form of an adjustable 20 ft to 40 ft wide weir-like notch that reconnects hydrologic flow between Pond A8 and Alviso Slough. Construction of the A8 notch is scheduled to begin in the fall of 2009 (C. Strong, pers. comm.). The concern surrounding breaching Pond A8 encompasses both the scour (due to increased tidal prism) and redistribution of sedimentary Hg in adjacent Alviso Slough (which has sediment total mercury (THg) concentrations 3-times higher than in the greater South Bay), and changes to MeHg dynamics within Pond A8 (Marvin-DiPasquale and Cox 2007, Grenier *et al*. 2007).

It is estimated that sediment scour in Alviso Slough with either of the notch scenarios will result in a substantial amount of Hg being remobilized to the surrounding environment (approximately 66 to 125 kg of THg, 0.05 to 0.10 kg of reactive inorganic mercury ($Hg(II)_R$), and 0.08 to 0.14 kg of MeHg; Marvin-DiPasquale and Cox 2007). It is not known how much of each Hg fraction will be transported to the larger South Bay, into Pond A8, or upstream of Pond A8. However, exposure of the buried (chemically reducing) sediment to oxygenated overlying slough water may result in significant changes in the speciation of remobilized Hg, potentially enhancing MeHg production. Specifically, concentrations of $Hg(II)_R$ increased 40-60X within 7 days when anoxic Alviso Slough sediment collected from a depth of 150 cm was mixed with oxygenated slough water (Figure 3). Since $Hg(II)_R$ is a surrogate measure of the fraction of total inorganic Hg(II) that is available for microbial conversion to MeHg, this finding has major implications about the potential for enhanced MeHg production in Pond A8 and the surrounding region.

Within Pond A8 itself, MeHg concentrations in the sediments and biota are among the highest of any measured in the entire South Bay (Miles and Ricca in press, Ackerman *et al.* 2007a,b, Ackerman and Eagles-Smith 2008, Grenier *et al.* 2007). Although, it is unclear how Hg cycling within the pond will change post-breach, other recently breached salt ponds in the region (A19 and A20) showed more than 5-fold increases in sediment MeHg concentrations post-breach

(Miles and Ricca, in press). Thus, there is the potential that MeHg concentrations within the pond may increase above the currently high levels.

Conversely, related studies (Grenier *et al.* 2007) suggested that the elevated microbial activity in Pond A8 relative to Alviso Slough is driven by high loading of readily degraded phytoplankton (Figure 4). This, coupled with the high $Hg(II)_R$ concentrations associated with Pond A8 flats may be responsible for significantly higher %MeHg in Pond A8 than in Alviso Slough (Figure 5). This study concluded that returning Pond A8 to tidal flushing will likely decrease net MeHg production within the footprint of Pond A8, as phytoplankton densities in the overlying water, and the subsequent deposition of phytoplankton to the benthos (which likely fuels the methylation process) would be reduced relative to its current status.

In a more recent study comparing Pond A11 (low in phytoplankton) with Pond A12 (high in phytoplankton), dissolved and particulate MeHg concentrations were significantly elevated in Pond A12 water, as was the %MeHg associated with the particulate fraction (Figure 6). Biosentinel fish further indicated that bioaccumulation of MeHg was significantly higher in Pond A12 (Figure 7). These data support the hypothesis that more organic production in the form of phytoplankton leads to more MeHg formation and bioaccumulation. Thus, for breached ponds such as A8, resulting changes in hydrology and primary productivity within the ponds may substantially alter MeHg bioaccumulation in biota that forage there.

Although the Alviso Pond/Slough Complex contains more THg than other areas of the South Bay (SFEI 2005, Marvin-DiPasquale and Cox 2007), wetland restoration may not necessarily increase MeHg in the local food web because MeHg production depends on many environmental factors in addition to THg concentration. Recent studies indicate significant spatial variation in Hg bioaccumulation are related to differences in habitat type (Eagles-Smith *et al.* 2008, 2009). Even within a single type of wetland, Hg bioaccumulation within the same biosentinel species can vary greatly among wetlands with different characteristics (Grenier *et al.* 2007). Further, Hg concentrations in several waterbird species vary greatly even among adjacent wetlands (Ackerman *et al.* 2007a,b, 2008a,b,c), indicating the importance of processes governing MeHg production, transport and partitioning (among solid and dissolved phases) that occur within wetlands. In order to understand how management actions influence MeHg production and bioaccumulation into the food web, an integrated monitoring program that incorporates abiotic and process studies with biological indicators of exposure is recommended (Evers *et al.* in press). No single biosentinel species can provide the information needed across all habitats, spatial scales, and components of the food web. Thus, a multiple biosentinel approach is proposed to determine how management actions will affect MeHg in the food web, and ultimately risk to sensitive wildlife.

B. The Biosentinel Toolbox

The biosentinel approach is based on developing appropriate biological indicators of Hg contamination that are indicative of local conditions over a relatively discrete spatial area and time frame, and that incorporate potential effects to at-risk species. However, most species do not occur widely across different habitats, and Hg availability can differ substantially among habitats within the same geographic area (Eagles-Smith *et al.* 2008, 2009, Ackerman *et al.* 2007a, b). Thus, no single biosentinel can provide managers with the information they need about where and when their management actions are impacting Hg in the food web. An integrated monitoring program that incorporates multiple biosentinels is needed. Our approach in this proposal builds on a compilation of several years of research in the South Bay Restoration

Project Area, as well in the greater Estuary, and has focused on biosentinel development and appropriate scales of implementation. In addition, recent research on toxicological thresholds of Hg impairment to avian reproduction for waterbirds in the region (Ackerman and Eagles-Smith 2008, Eagles-Smith and Ackerman 2008) will provide benchmark values to assess potential risk and effects of restoration on sensitive wildlife.

STUDY OBJECTIVES:

Wetland restoration and management practices that would minimize MeHg bioaccumulation are not well known. Therefore, this proposal aims to monitor changes in Hg bioaccumulation that may occur after the planned breach of Pond A8, which will return it to muted tidal action. Biosentinel monitoring will be coupled with water and sediment chemistry to understand the processes that cause changes in Hg bioaccumulation and to determine if and how the operation of the A8 Notch causes a direct change in MeHg production in Pond A8 or in Alviso Slough. An increase in the bioavailability of MeHg could negatively impact breeding waterbirds, a result opposite to the management goal of restoring waterbird habitat for the Don Edwards San Francisco Bay National Wildlife Refuge and the SBSP Restoration Project. An increase in MeHg export to surrounding waters, habitats, and the wider Bay also could have important regulatory ramifications. By monitoring across multiple habitats and spatial scales, we will increase the information that managers can draw upon as they attempt to minimize Hg risk while moving forward with restoration. As such, the primary objectives of this proposal are to:

- Assess the impact of the A8 notch on Hg cycling within Pond A8 and Alviso Slough main-channel and adjacent marshes using an integrated biosentinel approach coupled with a stable isotope food web assessment, and process-level water-column and sediment studies.
- Determine the extent of the effect of the A8 notch implementation over time and with distance from the restoration site, and the relative effect among the different habitats and biosentinel species.
- Use water column and sediment mercury concentration and speciation data to link the underlying processes of MeHg production to bioaccumulation, and to investigate whether MeHg production potential changes as a function of changes in a) phytoplankton production, and/or b) Hg remobilization associated with Alviso Slough sediment scour.

STUDY AREA:

The primary study area will be within the Don Edwards San Francisco Bay National Wildlife Refuge and focused on Pond A8, Alviso Slough, and adjacent salt ponds, sloughs and marshes (Figure 8). We also will monitor appropriate control sites, including two ponds and one slough habitat. Control ponds will include one positive control (previously breached pond that interacts hydrologically with the adjacent slough) and one negative control (unbreached salt pond). A16 and A3N were selected as the positive and negative control ponds, respectively, and Mallard Slough was selected as the control slough. Control sites will be critical to assess baseline Hg bioaccumulation that is not associated with the opening of Pond A8 for three reasons. First, Pond A16, A3N and Mallard Slough are configured similarly to Pond A8 and Alviso Slough in that Pond A16 was recently opened to Mallard Slough and A3N is managed as a seasonal pond similarly to A8. Second, Pond A16, A3N and Mallard Slough are hydrologically separated from Pond A8 and Alviso Slough, so there will be no carryover effects. Finally, data collected in Pond

A16 will provide useful baseline data for when Pond A16 is enhanced by creating additional waterbird nesting islands, currently scheduled for construction in 2011 at the earliest.

APPROACH:

To monitor the effect of the Pond A8 notch on MeHg bioaccumulation within the pond and the surrounding environment, we have identified four biosentinels that fall into two groups: waterbird eggs and small fish. These biosentinels will provide important information on Hg bioaccumulation within specific habitats and locations, as well as allow managers to evaluate overall changes in Hg-related wildlife risk. The waterbird group provides pond-specific information on Hg bioaccumulation from both invertebrate (avocets) and fish-based (terns) prey, and is a precise indicator of potential risk to wildlife reproductive impairment (Figure 9). The fish are localized populations that provide comparative information on Hg availability within the same matrix over time and across habitats. Below are the four individual biosentinels that comprise these groupings.

1. **Forster's Terns (*Sterna forsteri*)** are fish-eating birds that nest in high densities at multiple sites within the South Bay Salt Ponds (Strong *et al.* 2004) and forage in salt ponds and adjacent marshes (Ackerman *et al.* 2008a). As top predators, changes in MeHg bioavailability in the system are amplified in their tissues relative to lower trophic level species. Previous research has shown that terns have substantially higher Hg levels than any of the 13 bird species sampled in the Bay to date (Figure 10), and nearly half of all tern eggs sampled in the South Bay exceed known toxicological thresholds (Ackerman and Eagles-Smith 2008). Moreover, once Forster's terns arrive in the South Bay to breed, they have relatively small space use (Ackerman *et al.* 2008b, Bluso-Demers *et al.* 2008). Any changes in MeHg production associated with A8 habitat restoration will likely occur within the A8 complex and the adjacent wetlands where terns forage (Ackerman *et al.* 2008b). Therefore, monitoring tern eggs provides important information on how wetland management practices may alter overall risk of Hg exposure to wildlife.

2. **American Avocets (*Recurvirostra americana*)** are invertebrate-foraging shorebirds that are abundant in the region year-round and are the most abundant breeding shorebird in San Francisco Bay (Stenzel *et al.* 2002, Rintoul *et al.* 2003). Recent radio telemetry studies (Ackerman *et al.* 2007a, Demers *et al.* 2008) have shown that during the eight weeks approaching egg laying, avocet space use is highly localized and occurs predominantly within the ponds where nesting occurs. Thus, avocets are excellent indicators of Hg concentrations in the invertebrate food web at the "individual-pond" spatial scale. Avocets nest at high densities across a wide range of habitats, including salt pond islands, dried salt pond pannes, and vegetated marshes, highlighting their utility across the entire SBSP Restoration Project area (Ackerman *et al.* 2006). Hg concentrations in avocet eggs (which are reflective of diet only a few weeks prior to laying) differ widely among colonies. In fact, differences between nearby colonies can differ by up to a factor of 5, indicating their utility as Hg biosentinels at a small spatial scale (Figure 11).

3. **Threespine stickleback (*Gasterosteus aculeatus*)** are a small fish species with well-studied behavior and ecology that occurs widely throughout the restoration area, is strongly linked with water column prey, and which represents an extremely important conduit for Hg transfer through the food web (Eagles-Smith and Ackerman, *in press*). These fish are short-lived (1-yr) and are found in loosely aggregated shoals. Additionally, USGS BRD has an extensive stickleback database (2005-2008) with Hg concentrations throughout the SBSP Restoration Project area ponds which will provide baseline conditions before extensive restoration occurs in the Alviso Salt Pond Complex (Figures 12, 13). This biosentinel will be used to assess changes in Hg bioaccumulation within

ponds and adjacent sloughs, thus allowing for comparisons of changes in Hg bioaccumulation with time and between habitats.

4. **Mississippi silverside (*Menidia audens*)** is a small fish species that provides a food-web linkage from the sloughs to the wider South Bay. This species has been developed as a highly effective spatial and temporal biosentinel of MeHg exposure throughout the Bay-Delta, particularly in relation to TMDL regulatory considerations (Figure 14, Slotton *et al.* 2002, 2007). Silversides are an abundant and important prey species in the sloughs and Bay margins, with a wealth of comparative data (Greenfield *et al.* 2006, Slotton et al 2008, Figure 15). Moreover, silversides are relatively localized, and show rapid response to changes in Hg availability.

Stable Isotopes

Although MeHg concentrations in biota generally increase at higher trophic levels (Wiener *et al.* 2007), recent research has shown that foraging habitat is another critical determinant of MeHg exposure in biota (Eagles-Smith *et al.* 2008, 2009). Thus, two organisms occupying identical trophic positions can have quite different MeHg concentrations due to differences in foraging habitats. Moreover, restoration activities will likely alter the community structure within the SBSPRP area, resulting in changes in the food web that can ultimately influence biotic Hg concentrations. Thus, any changes in biosentinel Hg concentrations may not be attributable to specific mechanisms, whether they are shifts in the food web or altered MeHg production. To address this we will evaluate potential food web mediated changes in Hg bioaccumulation through the use of stable carbon, nitrogen and sulfur isotopes.

Stable isotopes provide a powerful and cost-effective tool for assessing the trophic linkages within food webs, and when coupled with Hg analyses can provide important information on how MeHg flows through food webs. Although isotopes do not provide taxonomic resolution in feeding linkages, they allow for a quantitative assessment of foraging habitat (e.g. benthic vs. pelagic; $\delta^{13}C$), trophic position ($\delta^{15}N$), and biogeochemistry (e.g. sulfate reduction; $\delta^{34}S$) that may drive MeHg bioaccumulation. They will also be critical for determining whether any measured changes in biosentinel Hg levels are the result of altered MeHg production or trophic relationships.

METHODS:

<u>Methods: Waterbird egg sampling (USGS-BRD):</u> We will monitor Hg concentrations in randomly collected avocet and Forster's terns eggs at 4 colonies per species located in the South Bay. Colony locations will depend on breeding conditions, however stable breeding colonies have occurred in Ponds A1, AB1, A7, A8, A16, R1, N4, New Chicago Marsh, and Eden Landing. These colonies represent a gradient of distances from the A8 (including A5/A7) wetland restoration complex and will be used to assess any changes in waterbird Hg bioaccumulation associated with restoration actions, and in relation to regional changes. To assess Hg concentrations in waterbird eggs, we will randomly sample one egg from up to 15 nests per colony for each species during 2010 and 2011 breeding seasons. We will refrigerate collected eggs until processing (<2 weeks), measure egg size and volume, then open each egg, remove all egg contents into a polypropylene jar, evaluate embryos for gross abnormalities and malpositions that can be caused by contaminants, and freeze the egg until THg analysis at the USGS Davis Field Station Hg Lab with Milestone DMA 80 Hg analyzer following EPA method

7473 (US EPA 2000). To test for changes in egg THg concentrations associated with restoration actions in the Pond A8 complex, we will use a residual-based analysis incorporating the data collected by USGS over the past 5 years (Ackerman *et al.* 2007b, Ackerman and Eagles-Smith 2008). More specifically, we will standardize relative egg Hg concentrations for each colony across years, and compare relative Hg levels in eggs sampled from colonies within or directly adjacent to A8 with concentrations in other colonies in the region that are not influenced by the A8 Hg signal. We will then test whether standardized egg Hg concentrations at each colony increase or decrease after the A8 management action.

<u>Methods: Pond fish sampling (USGS-BRD):</u> To assess the degree to which MeHg cycling is altered post-notch within Pond A8, we will use a resident, localized fish, the threespine stickleback, as our fish biosentinel. We will sample stickleback (N=10 per location and sampling event) at three locations in the Pond A8 complex (1 near the A8 notch and 2 elsewhere in the pond) at least three times per year, as well as twice bounding the breaching of the pond to Alviso Slough. We will also sample in A16 and A3N reference ponds to control for changes in ambient concentrations. Both ponds are strong candidates for reference sites because USGS has been monitoring Hg in stickleback between 2005 and 2008. Additionally, A16 was a former salt pond that has been subsequently linked to Mallard Slough, and is scheduled to undergo future pond restoration and enhancement as part of the SBSPRP. We will sample fish using standard methods such as beach seines and minnow traps. Fish will be cleaned and stored frozen at -20°C until THg analysis at the USGS Davis Field Station Hg Lab.

<u>Methods: Slough fish sampling (UC Davis):</u> This component of the project provides a linkage between the restoration activities at Pond A8 and the potential export of altered MeHg exposure conditions to surrounding waters. For this project, two small fish species will be used in the slough environment. Threespine stickleback will be taken in groups of 10 replicate individuals, each to be analyzed separately, from a control slough (Mallard Slough) and 4 strategically located sites along Alviso Slough: upstream of the notch, at the notch, midway down the slough, and near its base. A second species, Mississippi silverside, will be used as a more widely integrative biosentinel, providing a linkage to the Bay and to a large amount of comparative data from locations throughout the Bay-Delta watershed. Silversides are targeted for collection at all of the sites listed above, except the uppermost site on Alviso Slough, which is normally above their range. Composite samples will be used, with 6 replicate composites of up to 10 fish per composite, to reduce budget costs and provide data comparable to other regionally collected fish data (RMP). Three seasonal collections will be made in each of two years, plus two additional samplings bracketing the main notch-opening event. We will use well-established protocols to sample slough fish with seines (Slotton et al 2007), and preserve fish frozen prior to analysis. Analyses will be performed using a Perkin-Elmer FIMS cold vapor atomic absorption system outfitted with an auto sampler unit.

<u>Methods: Stable Isotopes (USGS-BRD):</u> Fish collection will occur as described above. Additionally, we will sample a small number of obligate primary consumers (invertebrates) from each site in order to "baseline correct" for variance in ambient isotope ratios among locations. Samples will be processed as described in Eagles-Smith et al. (2008a), and after processing, samples will be measured into tin capsules and analyzed for stable isotope ratios via mass spectroscopy. We will use multi-source mixing models (i.e. IsoSource, IsoError) to quantify

energy sources to each fish, and compare trophic linkages and habitat-specific foraging with Hg concentrations before, during, and after the breach. This approach will allow us to determine the extent to which alterations in the aquatic food web influenced Hg bioaccumulation relative to alterations in MeHg production.

Methods: Water Column Mercury Dynamics (USGS-WRD): Biosentinel data are important for answering if, when, and where MeHg bioaccumulation is impacted by the hydrological changes from the construction and operation of the Pond A8 Notch (or similar management actions). However, biosentinel data alone do not offer an explanation as to why and how the observed changes in biota Hg concentrations occurred. Thus, process-level information regarding changes in Hg concentration and speciation in abiotic matrices (i.e. water) is essential to fully understand what the impact of a given management action was. This information also enables prediction of how MeHg concentrations in biota might be affected in the future, given similar or modified changes to the hydrology of managed ponds.

Spatial and temporal trends in water column Hg concentration and speciation will be assessed for both dissolved and particulate phases over the study period, and will be related to changes in the quality and quantity of suspended particulate material (i.e. phytoplankton and inorganic particles), dissolved nutrients (nitrate and phosphate), and dissolved organic carbon (DOC) and specific ultra-violet absorption (SUVA, and measure of organic matter quality and origin). Water samples will be collected using trace metal clean sampling techniques (USEPA, 1996), at nine locations per sampling event, on ten occasions (at approximately 2.5 month intervals) to capture the complete seasonal trends before, during and after the construction of the Pond A8 Notch (See Table 1). The distribution of sampling sites will match the biosentinel sampling and will include: Alviso Slough (3 sites), Pond A8 (3 sites), Mallard Slough (reference, 1 site), Pond A16 (reference, 1 site), and Pond A3N (reference, 1 site). Whole water samples will be held on ice, in the dark in acid cleaned glass bottles until further processing at the laboratory (within 24 hrs of field collection). Field measurements will include water column dissolved oxygen, temperature, pH and conductivity.

Further processing and sub-sampling in the laboratory will include the collection of the particulate phase on pre-combusted / pre-weighed glass fiber filters. Non-filter passing particulates will be collected for each of the following analytes: THg, MeHg, total suspended solids (TSS), particulate carbon and nitrogen (PC/PN), and chlorophyll. The filtrate will be sub-sampled for the following dissolved analytes: THg, MeHg, DOC and nutrients. All particulate samples will be preserved at -80 ^{o}C. Dissolved THg and MeHg samples will be analyzed on an Automated Hg Analyzer (Tekran Model 2600), according to EPA Method 1631 (USEPA, 2002). Particulate MeHg samples will be analyzed on a Brooks Rand automated MeHg analyzer, following Bloom (1989). Dissolved THg will be quantified on the Tekran Model 2600 Automated Hg Analyzer (USEPA, 2002). TSS filters will be freeze dried, then reweighed to calculate the mass per volume filtered. PC/PN filters will be first acid fumed in a dessicator to remove any carbonate minerals, and then will be analyzed on a Carla Erba 2500 elemental analyzer connected to an Elementar Isoprime mass spectrometer (Kendall *et al.* 2001).

Within 24 hours of collecting the filtrate the UV absorption of an aqueous sub-sample will be assessed spectrophotometrically at 254 nm wavelength using a Shimadzu Model UV-1601 spectrophotometer (Shimadzu Scientific Instruments). DOC samples will be preserved with 0.1% HCl and subsequently quantified via high temperature combustion and IR detection (Qian and Mopper, 1996) on a Total Organic Carbon Analyzer (Model TOC-VCPH, Shimadzu

Scientific Instruments). SUVA will subsequently be calculated from the UV absorption and the DOC concentration (USEPA 2005). Dissolved nutrient samples will be stored frozen and will be subsequently analyzed on an automated Aquakem 250 nutrient analyzer, according to manufactures recommendations.

Methods: Sediment Mercury Dynamics (USGS-WRD): Sediment collection and associated measurement will occur at all nine locations where water collections occur, sampling six times over two years (between the 2^{nd} quarter of Year-1 and the 4^{th} quarter of Year-2), and in conjunction with six of the ten water/fish sampling events. Surface sediment (top 0-2 cm) will be collected, from a boat or from shoreline access, as previously described (Grenier and others, 2009; Lutz and others, 2008). Sediment will held on ice in acid cleaned mason jars until further processing (within 24 hours) under anaerobic conditions at the USGS facility in Menlo Park, Calif. (Grenier and others, 2009; Marvin-DiPasquale and others, 2008).

Measurements of Hg biogeochemistry in surface sediment will include MeHg production rates (via stable isotope tracer ^{202}Hg(II) amendment**,** Hg-speciation (total-Hg, MeHg, and reactive Hg(II)), and a suite of key ancillary parameters known to impact Hg-cycling (sulfate reduction via radiotracer $^{35}SO_4^{2-}$ amendment, organic content, pH, redox, total reduced sulfur, solid phase iron (II and III) speciation, grain size and pore water sulfate). Due to budget constraints, microbial rate assays (MeHg production and sulfate reduction) will be limited to Pond A8, A3N and A16 sediment, while all other measurements will be conducted on sediment samples collected from all three Ponds, as well as from Alviso and Mallard Slough. Table 2 provides information on the purpose of each assay and the analytic methods to be used.

Methods - QA/QC: We will follow rigorous QA/QC protocols which include analysis of appropriate method blanks, certified reference materials, duplicates, and matrix spikes with each analytical batch.

Statistical analysis: We will use Generalized Linear Models (such as ANCOVA) to test whether Hg concentrations in biosentinels differed pre- and post-notch and relative to ambient changes over the same time period. We will develop a statistical model for each biosentinel that will include independent variables of site and year for all species and date, standard length, age and sex as appropriate. We will also include site × year interactions when possible to assess whether notch construction influenced Hg concentrations relative to any ambient change.

SYNERGIES WITH OTHER PRIORITY RESEARCH STUDY TOPICS:
Whereas this proposal addresses the entire Topic 2 “Assessment of Mercury Bioavailability Utilizing Sentinel Species,” it will also provide information relating directly to these other priority topics: Topic 5 “Pond, Slough, and Bay Water Quality Interactions”, Topic 3 “Waterbird Nesting and Foraging in Managed Ponds,” and Topic 7 “Effects of Restoration on Fish Assemblages.”

Together, these projects will provide a picture of where mudflat scour is occurring relative to where any effects of scour can be detected in biosentinels.

DATA ARCHIVING:
Data handling and storage will follow Federal Geographic Data Committee (FGDC) metadata standards. Primary data (field and laboratory) will be recorded in lab notebooks and on

standardized data collection forms or directly into a standardized data base using an Access form on a laptop computer. Field data will be referenced in GIS coverages, data projected in UTM in NAD83 horizontal and NAVD88 vertical datum. Data will be subsequently transferred into electronic workbooks (e.g. MS Excel, Access) for the purposes of calculation and database management. All data will be compiled, QA/QC checked, and archived on a data server with mirrored drives, tape backup, and redundant copies offsite. Both primary and electronic data will be preserved for a minimum of five years after the completion of the project. Datasets will be made available with permission for use specified in the metadata, and made accessible through the SBSPRP website. Biosentinel data will be integrated with the CALFED Bird Mercury Project, South Baylands Mercury Project, and UC Davis Mercury Biosentinel Program databases to provide standardization for comparisons over time and to facilitate future conversion to State formats (e.g. SWAMP).

WORK SCHEDULE:
Work will commence from final signature of the agreement for a period of two years (spanning parts of 3 calendar years) with an annual report delivered at the end of year one and a final report delivered at the end of year two. Most fieldwork will occur in Spring through Fall (see table below), but will bound the A8 Breach construction. Water sampling will occur at approximately 2.5 month intervals (see table below). Data analyses and report writing will occur during fall and winter of the second year, with a draft report due in March of the final year, and a final report delivered in May of the final year. Planning, field collections, data reduction, and write-up will all be coordinated among the collaborators.

Table 1. Project Timeline

Timeline by quarter	Year 1				Year 2				Year 3			
	1	2	3	4	1	2	3	4	1	2	3	4
Pre-project Planning												
Logistics/Coordination		x	x	x	x	x	x	x	x			
Field Collections												
Pond Birds (2×)		x	x			x	x					
Pond Fish (6×)		x	x	x		x	x	x				
Slough Fish (8×)		x	x	x		x	x	x				
Water (10×)		x	x	x	x	x	x	x				
Sediment (6×)		x	x	x		x	x	x				
Laboratory/Analytical			x	x	x	x	x	x	x			
Data Reduction/Archiving			x	x	x	x	x	x	x			
Report writing and review									x			

EXPECTED PRODUCTS:
Annual briefings and presentations will be provided to the Science Program and given at the South Bay Science Symposium. Annual progress reports and a final report will be delivered to the SBSP Restoration Project's Lead Scientist and Project Team. Additional presentations and scientific papers will be prepared for appropriate outlets. Expected journal paper topics include:

The effect of levee breaching on Hg cycling and bioaccumulation in the estuarine food web; How does returning former salt ponds to tidal action affect Hg in the surrounding ecosystems and their wildlife?

LITERATURE CITED:

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Table 2. Analytical methods summary for sediment and pore water samples. Replicates field and equipment blanks, certified reference material, matrix spikes and other QA/QC samples (as appropriate) will constitute approximately 10% of the samples analyzed for each parameter listed.

Matrix - Parameter	Purpose for Measurement	Analytical Method	Reference
Microbial MeHg Production	MeHg is the Hg species of primary concern for wildlife toxicity and is produced by bacteria in anoxic sediments. Identifying controls on, and minimizing, MeHg production in various habitats is a key component of both the SFB Hg Strategy and the SBSPRP.	$^{202}HgCl_2$ amendment of whole sediment samples → 4 hr. incubation → freeze kill → KOH/Methanol extraction of $CH_3{}^{202}Hg^+$ → quantification via ethylation, purge and trap followed by ICP-MS. Calculate from $^{202}Hg(II)$-methylation rate constant and native $Hg(II)_R$ concentration.	Sediment $^{202}Hg(II)$ incubation as per radiotracer $^{203}Hg(II)$ approach (Marvin-DiPasquale and others, 2008); MeHg extraction (Xianchao and others, 2003); MeHg quantification via ethylation, purge and trap (USEPA, 2001) and ICP-MS (Lambertsson and others, 2001); calculation as per Marvin-DiPasquale and others (2008).
Microbial Sulfate Reduction	SO_4^{2-} reducing bacteria produce MeHg. This microbial pathway also produces reduced-S end products that can mediate Hg(II) bioavailability	$^{35}SO_4^{2-}$ incubation → distillation and trapping of reduced ^{35}S species via Cr(III) reduction and H_2S volatilization with acid → reduced ^{35}S quantification via LSC. Calculated from radiotracer derived rate constant and pore water sulfate concentration.	Marvin-DiPasquale and others, 2008
Sediment – Total Mercury (HgT)	Mercury is a toxic element of concern that bioaccumulates in aquatic food webs.	Heated aqua regia (HNO_3/HCl) sample digestion → oxidation to Hg(II) with BrCl→ reduction to elemental Hg^0 with $SnCl_2$ → trapped on gold covered sand → quanitified via automated CVAFS	USEPA, 2002; as modified by Olund and others (2004)
Sediment – Methylmercury (**MeHg**)	The form of mercury, which is most toxic, which most readily bioaccumulates up aquatic food webs, and which is produced by bacteria in sediments.	MeHg extraction via KOH/Methanol → aqueous phase ethylation → GC separation → pyrolization → quantification via CVAFS	Xianchao and others, 2003

Matrix - Parameter	Purpose for Measurement	Analytical Method	Reference
Sediment – Inorganic reactive mercury ($Hg(II)_R$)	The operationally defined pool of mercury that is a surrogate measure of the Hg(II) most readily available to bacteria for methylation. Value need for calculation of MeHg production rates.	Anoxic transfer into N_2 purged bubbler containing 0.5% HCl → $SnCl_2$ reduction of $Hg(II)_R$ fraction during a 15 minute purge cycle → trapped on gold covered sand → quantified via CVAFS	Marvin-DiPasquale and Cox, 2007
Bulk density and porosity	Needed to normalize Hg species concentration and microbial rates to sediment dry weight and wet sediment volumes, respectively.	Calculated from sediment wet and dry weight determinations	Marvin-DiPasquale and others, 2008
Sediment - Redox Potential (E_h)	Sediment characterization parameter reflecting the ratio of all oxidized/reduced species. May influence Hg bioavailability to microbes	ORP Pt-electrode placed directly into sediment and left to equilibrate (ca. 5-10 minutes).	Marvin-DiPasquale and others, 2008
Sediment pH	Sediment characterization parameter. May influence Hg bioavailability to microbes and microbial activity.	pH electrode placed directly into sediment	Marvin-DiPasquale and others, 2008
Sediment - Total reduced sulfur (TRS)	Primarily dissolved H_2S and solid phase FeS, which may control the partitioning of Hg species between solid and dissolved pools within sediment	HCl dissolution of solid phase FeS → volatile H_2S trapping in Zn-acetate → quantification via the methylene blue colorimetric assay	Marvin-DiPasquale and others, 2008
Pore water - Sulfate: (SO_4^{2-})	Primary electron acceptor for microbial sulfate reduction. Needed to calculate in-situ rates of SO_4^{2-} reduction	Pore-water collection via centrifugation (anoxic) → pore-water filtration (0.45 µm) → quantification via ion chromatography	Marvin-DiPasquale and others, 2008

Matrix - Parameter	Purpose for Measurement	Analytical Method	Reference
Sediment – solid phase acid-extractable Ferrous Iron: $Fe(II)_{AE}$	The end-product of microbial Fe(III)-reduction, In conjunction with solid phase Fe(III) concentrations, $Fe(II)_{AE}$ can tell us something about where a given sediment sample is poised along a continuum of conditions, from those favoring microbial Fe(III) reduction to those favoring microbial sulfate reduction.	Weak acid extraction (0.5 M HCl for 30 min) → centrifugation and supernatant sub-sampling → quantification via the ferrozine colorimetric assay.	Lovley and Phillips (1986), as modified by Marvin-DiPasquale and others (2008)
Sediment – solid phase amorphous (poorly crystalline) Ferric Iron: $Fe(III)_a$	The form of Fe(III) most readily available for microbial Fe(III)-reduction. In conjunction with solid phase $Fe(II)_{AE}$ concentrations, Fe(III) can tell us something about where a given sediment sample is poised along a continuum of conditions, from those favoring microbial Fe(III) reduction to those favoring microbial sulfate reduction.	Initial steps outlined above for the measurement of solid phase $Fe(II)_{AE}$ → addition of hydroxylamine to convert any remaining Fe(III) to Fe(II) → remeasure Fe(II) via ferrozine colorimetric assay. Calculate by difference (subtracting initial $Fe(II)_{AE}$ concentration prior to hydroxylamine addition).	Lovley and Phillips (1987), as modified by Marvin-DiPasquale and others (2008)
Sediment – solid phase crystalline Ferric Iron: $Fe(III)_c$	The form of Fe(III) less available for microbial Fe(III)-reduction, compared to $Fe(II)_a$. In conjunction with solid phase $Fe(II)_{AE}$ concentrations, Fe(III) can tell us something about where a given sediment sample is poised along a continuum of conditions, from those favoring microbial Fe(III) reduction to those favoring microbial sulfate reduction.	Extraction in dithionite/citrate for 1 hour (reduces all forms of Fe(III) to Fe(II)) → centrifugation and supernatant sub-sampling → quantification Fe(II) via the ferrozine colorimetric assay. Calculate by subtracting above measurements of solid phase $Fe(II)_{AE}$ and $Fe(III)_a$.	Roden and Zachara, (1996), as modified by Marvin-DiPasquale and others (2008)

Matrix - Parameter	Purpose for Measurement	Analytical Method	Reference
Sediment – Grain Size	THg concentration typically increases as ediment particle size decreases, due to the increased surface area to volume ratio. THg concentration is one factor that impacts $Hg(II)_R$ concentration and thus MeHg production	Wet sieve sediment through a 63 micron mesh → dry both fractions (< and > 63 micron) overnight at 105°C → weight dry sediment. Calculate and report the percentage of total mass < 63 microns.	Matthes and others, 1992

QUALIFICATIONS (CVs FOLLOW FIGURES):
This proposal is a collaborative effort among four Principle Investigators (PIs), each of whom has been conducting extensive research and directing mercury monitoring programs throughout the San Francisco Estuary, and elsewhere. Each PI has a strong track record of successfully managing large projects that integrate mercury studies across multiple disciplines, and bringing together researchers from different agencies and institutions. Additionally, a particular strength of this proposal is the ability to leverage baseline data from each of the PIs to clearly demonstrate any changes in MeHg exposure as a result of the A8 management action.

The organizational structure of the research team will be designed such that each PI is responsible for all aspects of a particular component of the study, but that the entire team together will integrate their work into a cohesive body of knowledge. As such, each PI will also be responsible for the administrative aspects of their particular component, but a single PI will coordinate all groups together. Dr. Eagles-Smith (USGS-BRD) will serve as the primary PI and administrative lead for this project, and will manage the administrative responsibilities. Please see CVs for further details and project participation.

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BUDGET AND STAFF ALLOCATIONS:
The total cost of this project is $684,223 over a two year period, and we are requesting $519,712 to complete all tasks. Thus we are providing a roughly 25% cost-share, with in-kind contributions totaling more than $160,000. Our request includes funds for <8% time for overall project PIs. Funds for supplies include fish sampling gear, egg collection and processing supplies, lab consumables, and office supplies. Detailed budgets for the overall project and by individual team follow.

South Bay Salt Pond Restoration Project
Restoring the Wild Heart of the South Bay

South Bay Salt Pond Restoration Project
Selected Monitoring and Applied Studies

The Effects of Wetland Restoration on Mercury Bioaccumulation in the South Bay Salt Pond Restoration Project: Using the Biosentinel Toolbox to Monitor Change across Multiple Habitats and Spatial Scales

TOTAL PROJECT BUDGET (combining all 3 collaborating groups)

Timeframe: Febrauary 2010-February 2012 depending on contract approval

Budget Categories	Total Project Budget			Total Grant Request			Total Proposed From Other Sources *(please specify the source, if known)*
	Year 1	Year 2	TOTAL	Year 1	Year 2	TOTAL	
Labor *(Includes the large majority of associated project analytical work)*	$212,546	$210,034	**$422,580**	$182,620	$179,901	**$362,521**	*App. $150,000 in matching Labor funding; see individual sub-budgets for details.*
Consultant fees/ Contractual Services	$16,190	$14,190	**$30,380**	$16,190	$14,190	**$30,380**	
Travel	$5,450	$5,450	**$10,900**	$5,450	$5,450	**$10,900**	
Project specific equipment, supplies/materials	$9,574	$8,324	**$17,898**	$9,574	$8,324	**$17,898**	*Not including >$35,000 in available equipment from USGS and UC Davis.*
Overhead	$56,421	$56,044	**$112,465**	$49,205	$48,808	**$98,013**	*App. $13,000 in matching Overhead funding from USGS Menlo Park.*
Other: *Related RMP small fish biosentinel Hg work in the South Bay*	$45,000	$45,000	**$90,000**	$0	$0	**$0**	*The related SFEI RMP Small Fish Biosentinel Program includes app. $45,000/yr monitoring at other comparable South Bay sites.*
TOTAL	$345,181	$339,042	**$684,223**	$263,039	$256,673	**$519,712**	

* See sub-budgets for details of each project component.
* Budget justification text included in main proposal document.

Sub-Budget 1
US Geological Survey-BRD; Waterbird and pond fish biosentinels; Project lead

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Sub-Budget 1 of 3 - USGS WERC Davis Field Station (pond bird and fish biosentinels)

Timeframe: February 2010-February 2012 depending on contract approval

Budget Categories	Total Project Budget			Total Grant Request			Total Proposed From Other Sources (please specify the source, if known)
	Year 1	Year 2	TOTAL	Year 1	Year 2	TOTAL	
Labor-Salaries and Benefits (agency: annual %FTE requested)							
Eagles-Smith (USGS: 9%/6%)	*$11,778*	*$9,040*	*$20,818*	*$8,852*	*$6,020*	*$14,872*	*$5,946 USGS contribution*
Ackerman (USGS: 4%)	*$7,182*	*$7,408*	*$14,590*	*$3,591*	*$3,704*	*$7,295*	*$7,295 USGS contribution*
Field/Lab Technician (USGS: 48%)	*$52,629*	*$52,629*	*$105,258*	*$52,629*	*$52,629*	*$105,258*	
Analytical (660 Hg samples)	*$28,050*	*$28,050*	*$56,100*	*$28,050*	*$28,050*	*$56,100*	
Labor (total)	$99,639	$97,127	**$196,766**	$93,122	$90,403	**$183,525**	
Consultant fees/ Contractual Services	$16,190	$14,190	**$30,380**	$16,190	$14,190	**$30,380**	Analysis of MeHg in subset of fish ($2000) and stable isotope analyses ($28,380)
Travel	$3,500	$3,500	**$7,000**	$3,500	$3,500	**$7,000**	
Project specific equipment, supplies/materials	$2,500	$1,250	**$3,750**	$2,500	$1,250	**$3,750**	*>$15,000 in equipment and on-site facilities by USGS*
Overhead	$10,564	$10,188	**$20,752**	$9,912	$9,515	**$19,428**	
Other:	$0	$0	**$0**	$0	$0	**$0**	
TOTAL	$132,393	$126,255	**$258,648**	$125,224	$118,858	**$244,083**	

* Budget justification text included in main proposal document.

Budget Justification: USGS BRD is tasked with monitoring mercury bioaccumulation in 3 of the 4 biosentinels, conducting stable isotope analysis, is responsible for developing the Quality Assurance Project Plan (QAPP) and is the overall administrative project lead that will be coordinating the project. We are requesting funds for <10% time for Project PIs. Additional PI time will be included as matching funds totaling $13,241. We are requesting funds for a technician's salary at 48% per year to conduct field work, sample processing, and data entry. Funds for supplies include consumables such as fish sampling gear, egg collection and processing supplies, lab consumables, and office supplies. USGS will provide equipment such as computers, boats, motors, and analytical equipment. All salaries include benefits and administrative costs.

Sub-Budget 2
UC Davis; Slough fish biosentinels

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Sub-Budget 2 of 3 - UC Davis (slough fish biosentinels)

Timeframe: Febrauary 2010-February 2012 depending on contract approval

Budget Categories	Total Project Budget			Total Grant Request			Total Proposed From Other Sources *(please specify the source, if known)*
	Year 1	Year 2	TOTAL	Year 1	Year 2	TOTAL	
Labor-Salaries and Benefits (agency: annual %FTE							
Slotton (UCD: 12%)	*$17,324*	*$17,324*	*$34,648*	*$10,269*	*$10,269*	*$20,538*	*Est. $14,110 UC Davis contribution*
Research Assoc. 1 (UCD: 16%)	*$12,685*	*$12,685*	*$25,370*	*$11,276*	*$11,276*	*$22,552*	*Est. $2,818 UC Davis contribution*
Research Assoc. 2 (UCD: 15%)	*$12,214*	*$12,214*	*$24,428*	*$9,178*	*$9,178*	*$18,356*	*Est. $6,072 UC Davis contribution*
Labor (total) *(includes Hg analyses and sample prep. for an estimated 592 samples)*	$42,223	$42,223	**$84,446**	$30,723	$30,723	**$61,446**	
Consultant fees/ Contractual Services	$0	$0	**$0**	$0	$0	**$0**	
Travel	$1,200	$1,200	**$2,400**	$1,200	$1,200	**$2,400**	
Project specific equipment, supplies/materials	$1,350	$1,350	**$2,700**	$1,350	$1,350	**$2,700**	*>$20,000 in equipment from UC Davis.*
Overhead (10%)	$3,327	$3,327	**$6,655**	$3,327	$3,327	**$6,655**	
Other: *Related RMP small fish biosentinel Hg work in the South Bay*	*$45,000*	*$45,000*	***$90,000***	$0	$0	**$0**	*The related SFEI RMP Small Fish Biosentinel Program includes app. $45,000/yr monitoring at other comparable South Bay sites.*
TOTAL	$93,100	$93,100	**$186,201**	$36,600	$36,600	**$73,201**	

* Budget justification text included in main proposal document.

Budget Justification: UC Davis labor and supply costs include mercury analytical and sample processing for an estimated 592 slough fish biosentinel samples. It is estimated that app. $23,000 will be contributed from other University funding toward planning, interpretation and writeup tasks not covered by the submitted budget. The small fish slough biosentinels will be comparable to and complemented by an extensive small fish data set from the Regional Monitoring Program (SFEI) at a cost of 150K/yr in total and app. 45K/yr specifically for the South Bay. This data set provides a regional context and ambient comparison for the project slough fish samples. The RMP small fish project thus represents and in-kind contribution of approximately $90,000.

Sub-Budget 3
U.S. Geological Survey – WRD Menlo Park; Water process studies

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Sub-Budget 3 of 3 - USGS Menlo Park (water process studies)

Timeframe: Febrauary 2010-February 2012 depending on contract approval

Budget Categories	Total Project Budget			Total Grant Request			Total Proposed From Other Sources (please specify the source, if known)
	Year 1	Year 2	TOTAL	Year 1	Year 2	TOTAL	
Labor-Salaries and Benefits (Pay grade, Hourly Rate; planned hrs/yr)							
J. Agee (GS-11; $45.90/hr; 192 hrs/yr)	*$15,513*	*$15,513*	*$31,026*	*$15,313*	*$15,313*	*$30,626*	
E. Kakouros (GS-11; $45.18/hr; 275 hrs/yr)	*$21,583*	*$21,583*	*$43,166*	*$21,583*	*$21,583*	*$43,166*	
L.H. Kieu (GS-9; $35.94/hr; 273 hrs/yr)	*$17,039*	*$17,039*	*$34,078*	*$17,039*	*$17,039*	*$34,078*	
Technician (GS-2; $11.65/hr; 140 hrs/yr)	*$2,840*	*$2,840*	*$5,680*	*$2,840*	*$2,840*	*$5,680*	
M. Marvin-DiPasquale (GS-14; $78.06/hr; 150 hrs/yr)	*$13,709*	*$13,709*	*$27,418*	*$2,000*	*$2,000*	*$4,000*	*Dr. Marvin-DiPasquale's time provided by USGS as a cost-share*
Labor (total) *(includes aqueous analytical work)*	$70,684	$70,684	**$141,368**	$58,775	$58,775	**$117,550**	
Consultant fees/ Contractual Services	$0	$0	**$0**	$0	$0	**$0**	
Travel	$750	$750	**$1,500**	$750	$750	**$1,500**	
Project specific equipment, supplies/materials	$5,724	$5,724	**$11,448**	$5,724	$5,724	**$11,448**	
Overhead (55.12% of NET)	$42,529	$42,529	**$85,059**	$35,965	$35,965	**$71,930**	
Other:	$0	$0	**$0**	$0	$0	**$0**	
TOTAL	$119,687	$119,687	**$239,375**	$101,214	$101,214	**$202,428**	

* Budget justification text included in main proposal document.

Budget Justification: Analytical costs for water column and sediment mercury speciation and SPM geochemical analyses, conducted by USGS (Menlo Park, CA), are reflected in staff salary plus benefits (90% of analytical costs) plus supplies (10% of analytical costs). Staff costs also includes those for data synthesis, interpretation, and report writing. All dollar amounts are given as gross costs using the current Federal government overhead rate of 55.12% of net costs, which includes both Bureau and Facilities (rent, operation and maintenance) costs. The budget for the work preformed by the USGS Menlo Park research group includes minimal salary for Dr. Mark Marvin-DiPasquale, which is projected at 300 hours over the course of the whole project, and equals an in-kind cost-share contribution of $23,418 net costs or $36,326 gross costs.

POTENTIAL REVIEWERS:
Chris Foe, Central Valley Regional Water Quality Control Board, Sacramento, CA;
email: cfoe@waterboards.ca.gov

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Chad Hammerschmidt, Wright State University; Dayton, OH;
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NECESSARY PERMITS:
Biosentinels will be monitored and handled under existing California Department of Fish and Game Scientific Collection (SC000009, SC801083-01, SC002545 and SC000084), Federal U.S. Fish and Wildlife Service (MB102896, TE-042630-3), and U.S. Geological Survey Bird Banding Laboratory (22911, 23446) permits, and Don Edwards San Francisco Bay National Wildlife Refuge Special Use Permit (11640-2006-006, 11640-2008-022, 81640-2008-060).

ANIMAL CARE AND USE:
All research will be conducted under approved study plans and guidelines of the U.S. Geological Survey, Western Ecological Research Center, Animal Care and Use Committee. This study proposes limited handling and disturbance of birds and is mainly observational in nature. Weekly nest monitoring activities have been extensively developed in cooperation with the Don Edwards SFBNWR staff over the past 5 breeding seasons and causes minimal disruption to nesting birds as indicated by low nest abandonment rates. Slough fish work by UC Davis will be conducted using UCD Animal Care and Use Protocol 13464, which was developed in conjunction with the UCD Wildlife Veterinary Unit to minimize any discomfort to sampled individuals, or disruption to sampled populations.

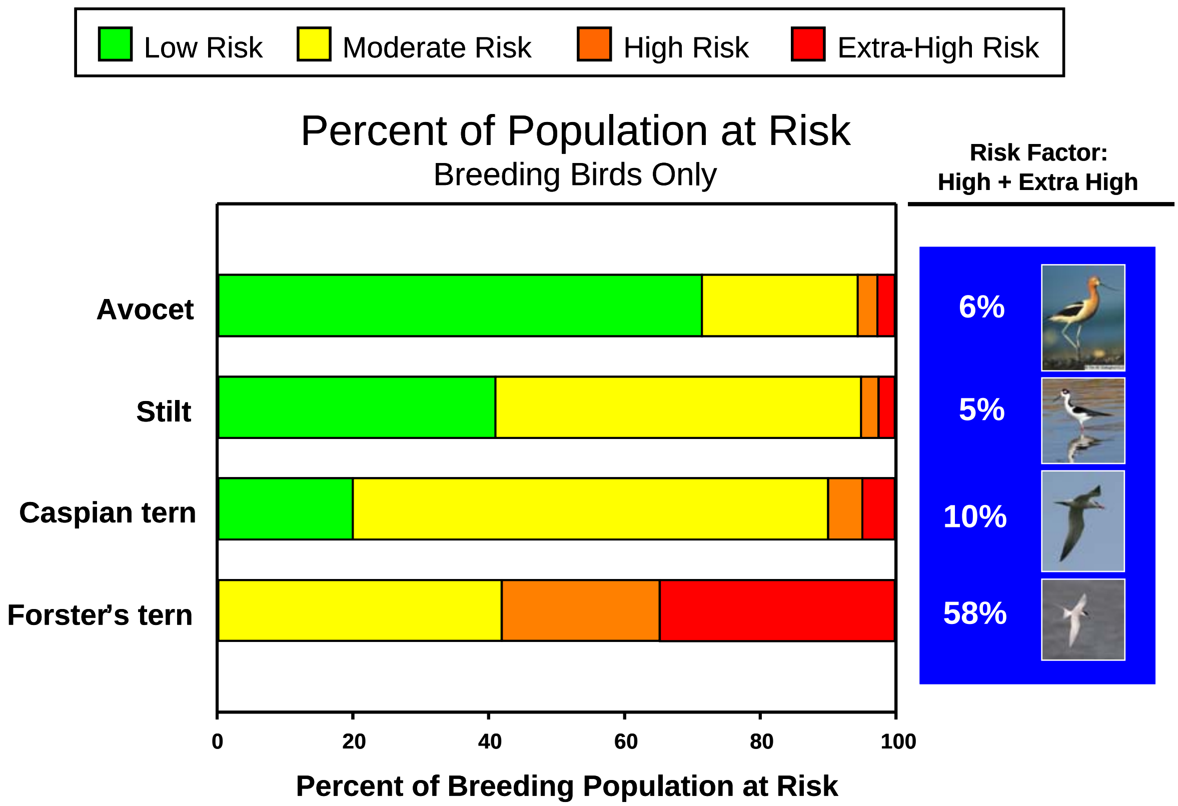


Figure 1. Percentage of waterbird breeding populations that are at risk to reduced reproductive success and declining populations due to mercury contamination in the South San Francisco Bay. Data are from birds captured on nests while incubating; blood was used as the sample matrix (Eagles-Smith et al. 2009). Risk levels were derived from Evers et al. 2008.

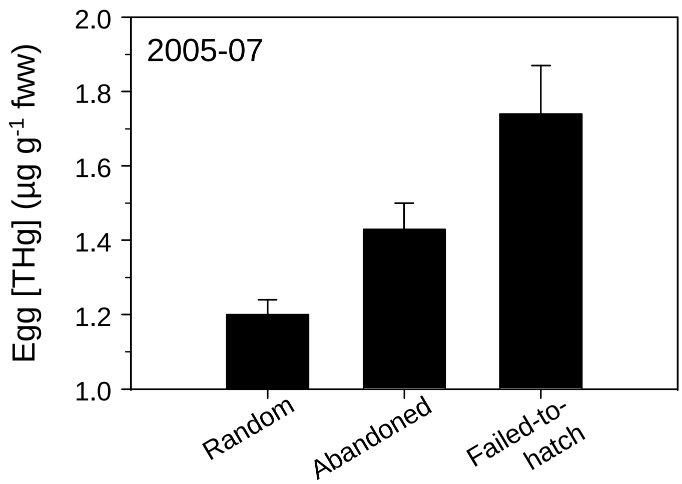


Figure 2. Mercury concentrations in failed-to-hatch Forster's Tern eggs were higher than randomly sampled eggs from successful nests during 2005-2007. Ackerman and Eagles-Smith 2008b.

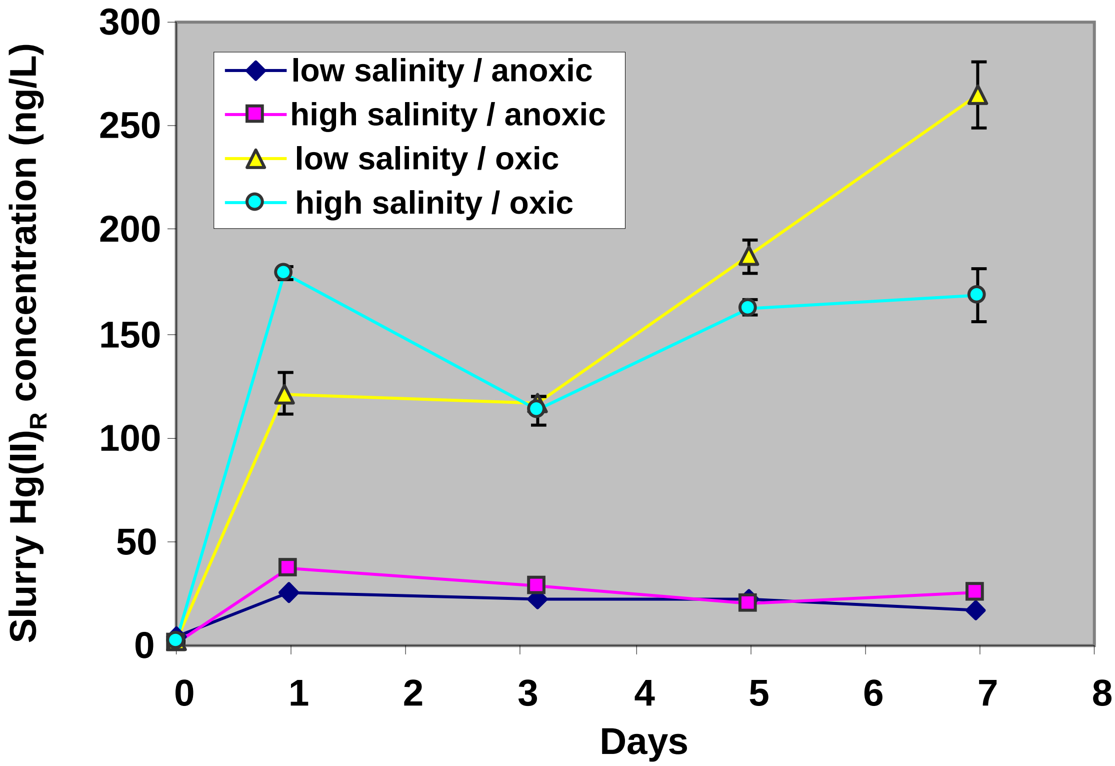


Figure 3. Time course plot of reactive mercury concentrations associated with the slough scour simulation experiment
Changes in reactive inorganic mercury ($Hg(II)_R$) concentrations in sediment / overlying slough water slurries repeatedly sampled over 7 days, under four treatment conditions (see inset legend). Error bars reflect the relative difference of n=2 sub-samples per treatment and time point. When not shown, error bars were smaller than the treatment symbol. Taken from Marvin-DiPasquale and Cox (2007).

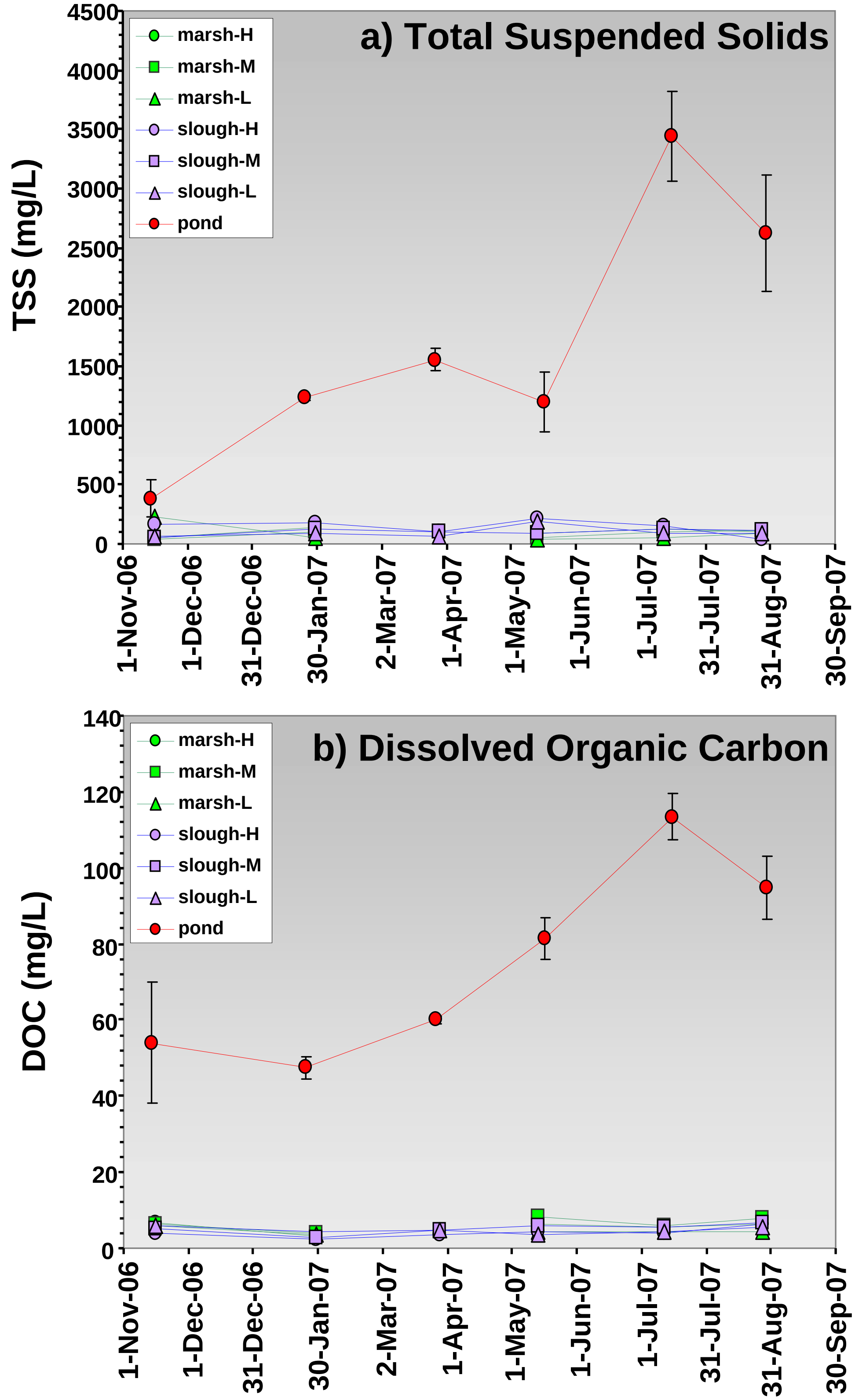


Figure 4. Time series graphs of overlying water concentrations of total suspended solids (a) and dissolved organic carbon (b), in waters collected from Alviso Marsh and Slough (high [H], mid [M] and low [L] along the salinity gradient), and Pond A8. Alviso Slough and Marsh symbols represent the average of N = 2 and N = 1 site(s), respectively. Pond A8 symbols represents the average of N = 7 sampling sites, and error bars represent ± 1 standard error of the mean. Visual examination of the samples indicates that the TSS in Pond A8 was almost completely phytoplankton. Taken from Grenier et al. (2008); unpublished.

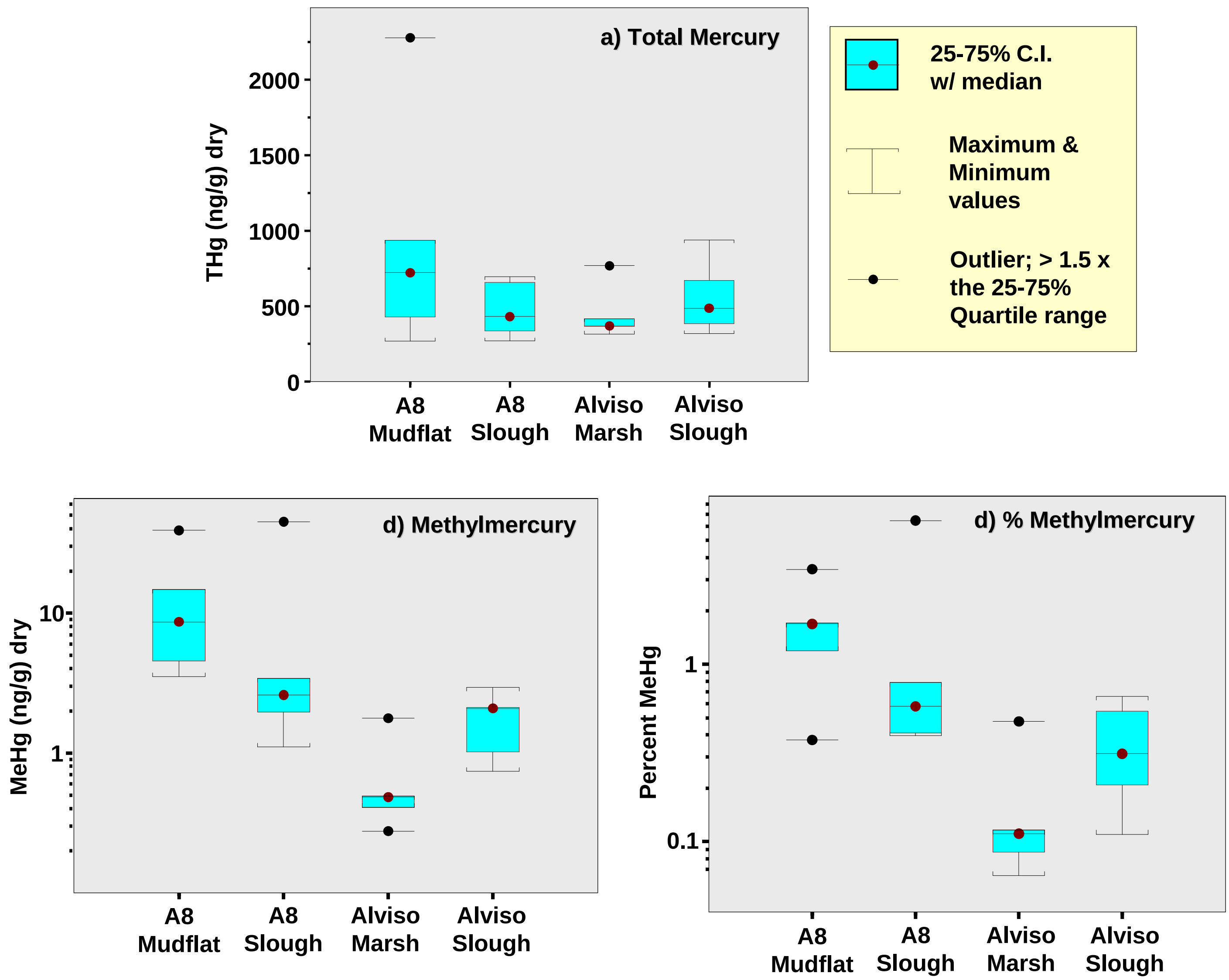


Figure 5. Box and whisker plots of sediment total mercury (THg), and methylmercury (MeHg) in Pond A8 and Alviso slough and Marsh, sampled between May and July 2007. Each habitat represents N = 5 individual sites. Taken from Greneir et al. (2008); unpublished.

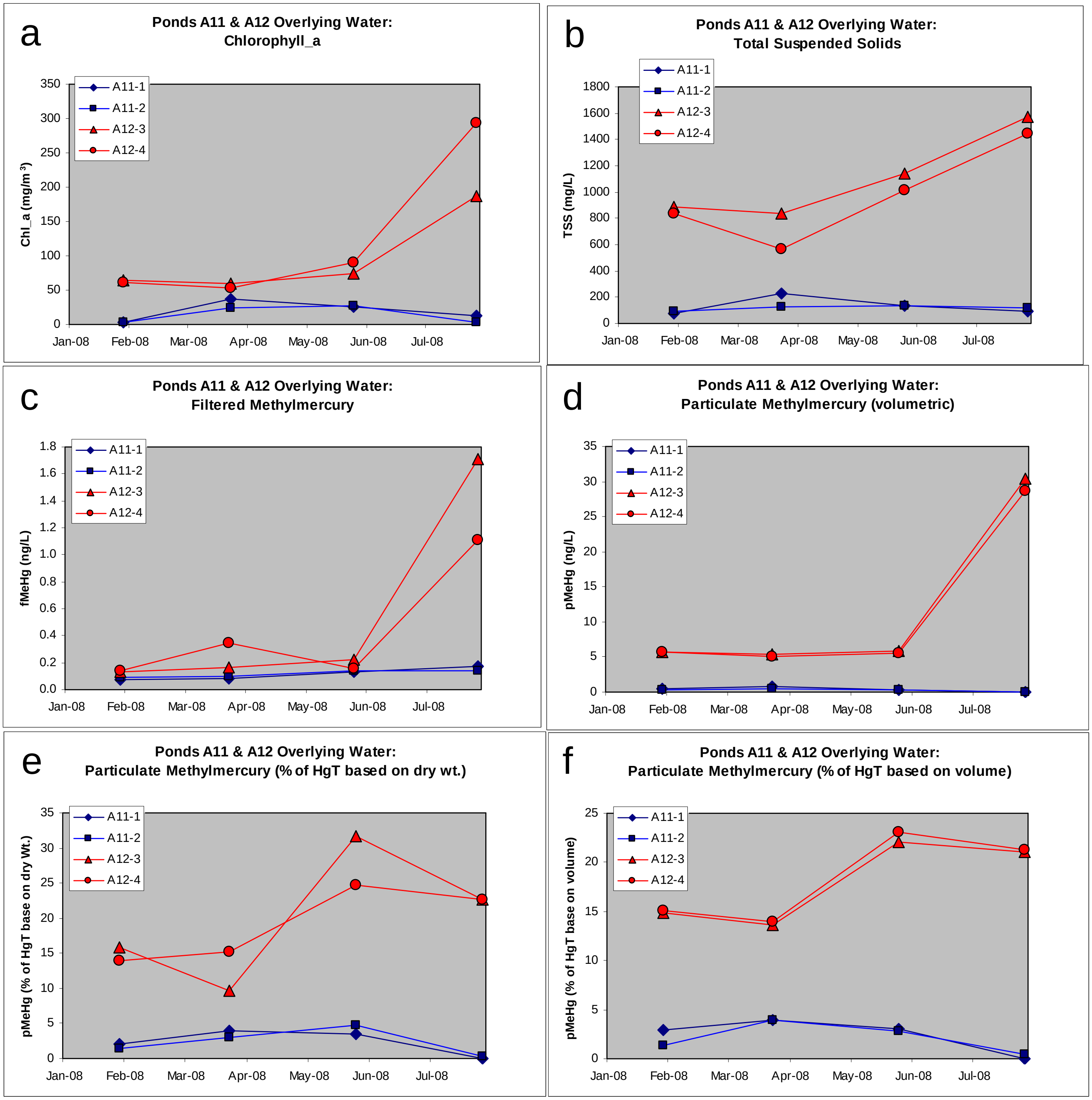


Figure 6. Time series graphs of overlying water concentrations of chlorophyll (a), total suspended solids (b) filtered MeHg (c), particulate MeHg (d), and percent MeHg on particles (e) and in whole water (f) in 2008 from two sites from Pond A11 and Pond A12. Marvin-DiPasquale, unpublished data.

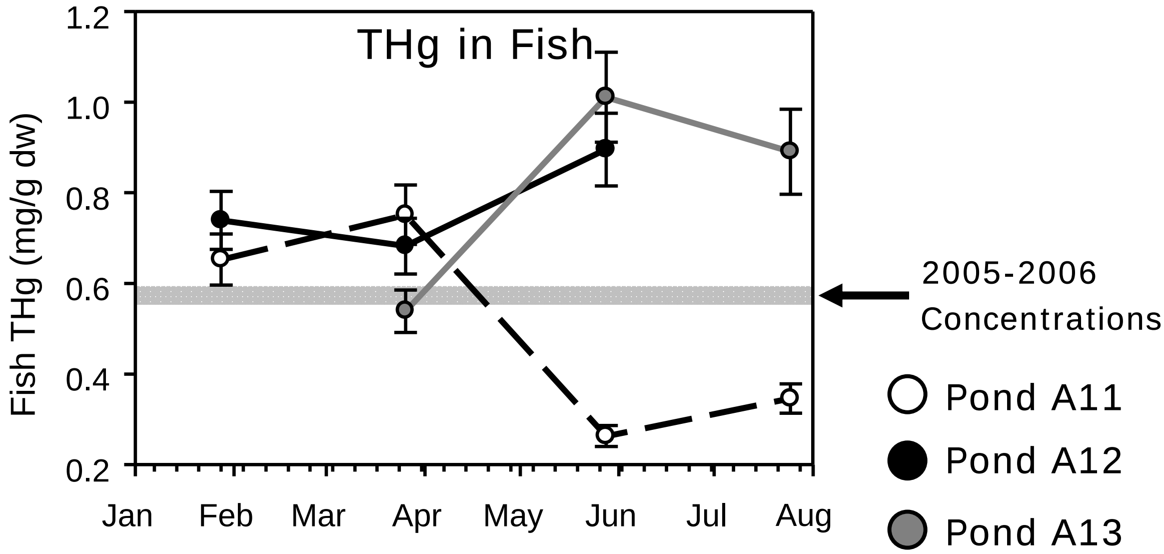


Figure 7. Temporal trend of THg concentrations in biosentinel fish from a reference pond (Pond A11), and pond undergoing management actions to increase waterbird nesting habitat (A12 and A13). Eagles-Smith and Ackerman, unpublished data.

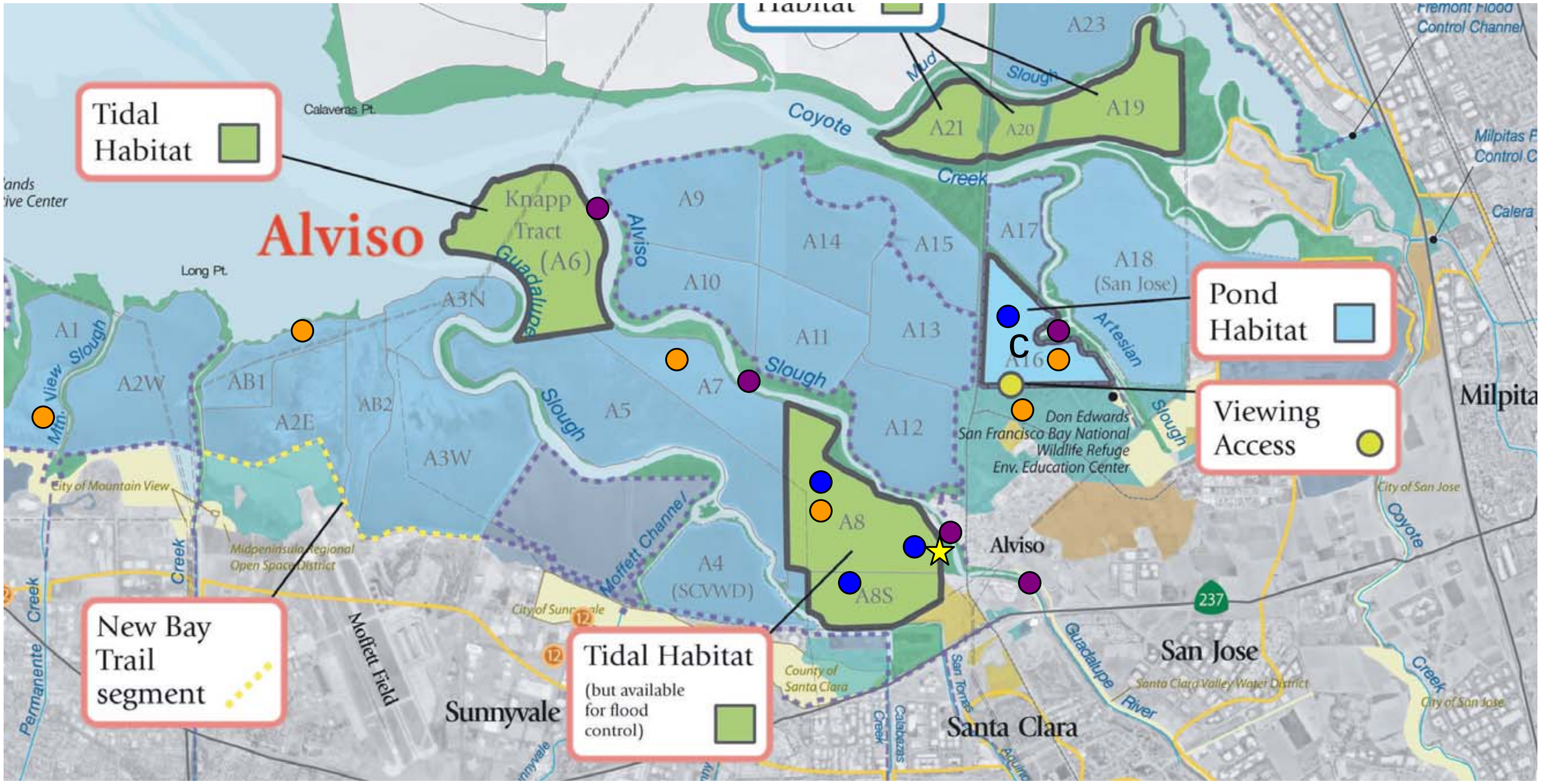


Figure 8. This example map shows how the study design relates to the geography of the SBSPRP. Orange circles indicate potential waterbird sampling sites representing a gradient of influence from Pond A8. Blue circles indicate example sampling location in ponds (Pond A8 and reference sites). Purple circles indicate example sampling locations in sloughs (Alviso Slough and reference site). Here, Pond A16, A3N and Mallard Slough are depicted as reference sites as an example, but the final choice is to be determined. The yellow star shows the approximate location of the future notch in the levee of Pond A8.
Map from www.southbayrestoration.org.

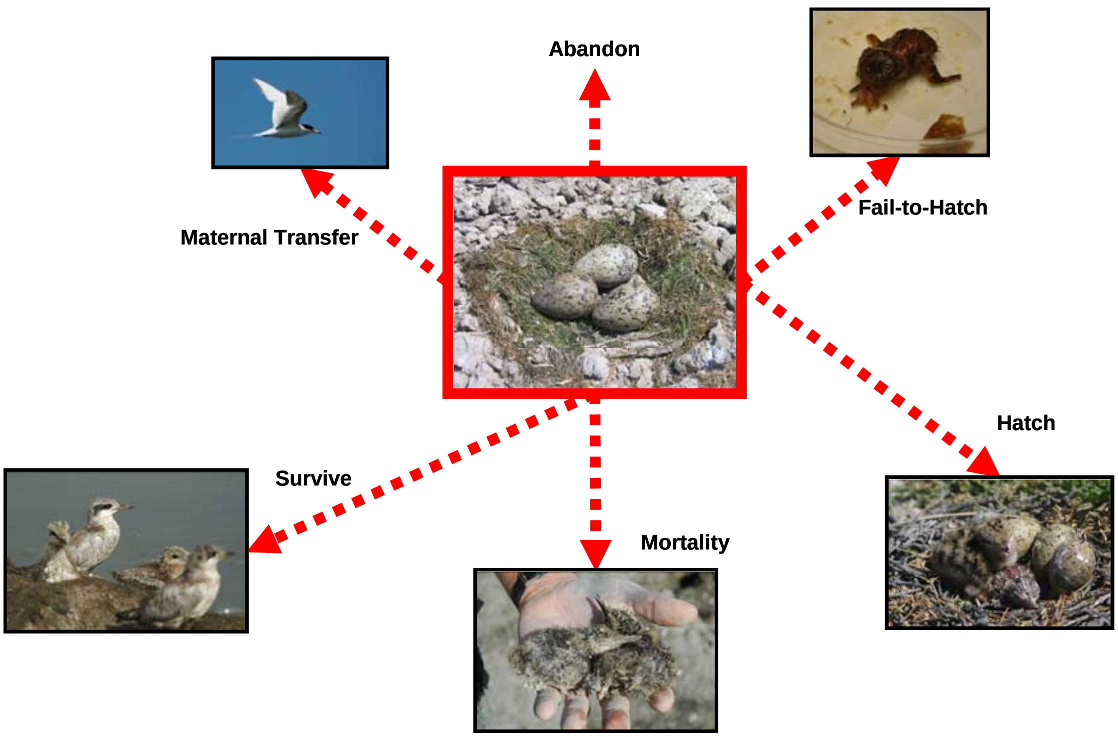


Figure 9. Conceptual model demonstrating the utility of using eggs as a monitoring tool for multiple lifestages, incorporating effects to adults, chicks, and eggs into a single tissue monitoring matrix – *eggs*. Once toxicity thresholds are developed for each lifestage shown, they can be translated into equivalent concentrations in eggs. Thereafter, toxicity thresholds for eggs will incorporate mercury's effect on hatchability, chick growth and survival, and the probability of adult nest abandonment. Ackerman and Eagles-Smith 2008b.

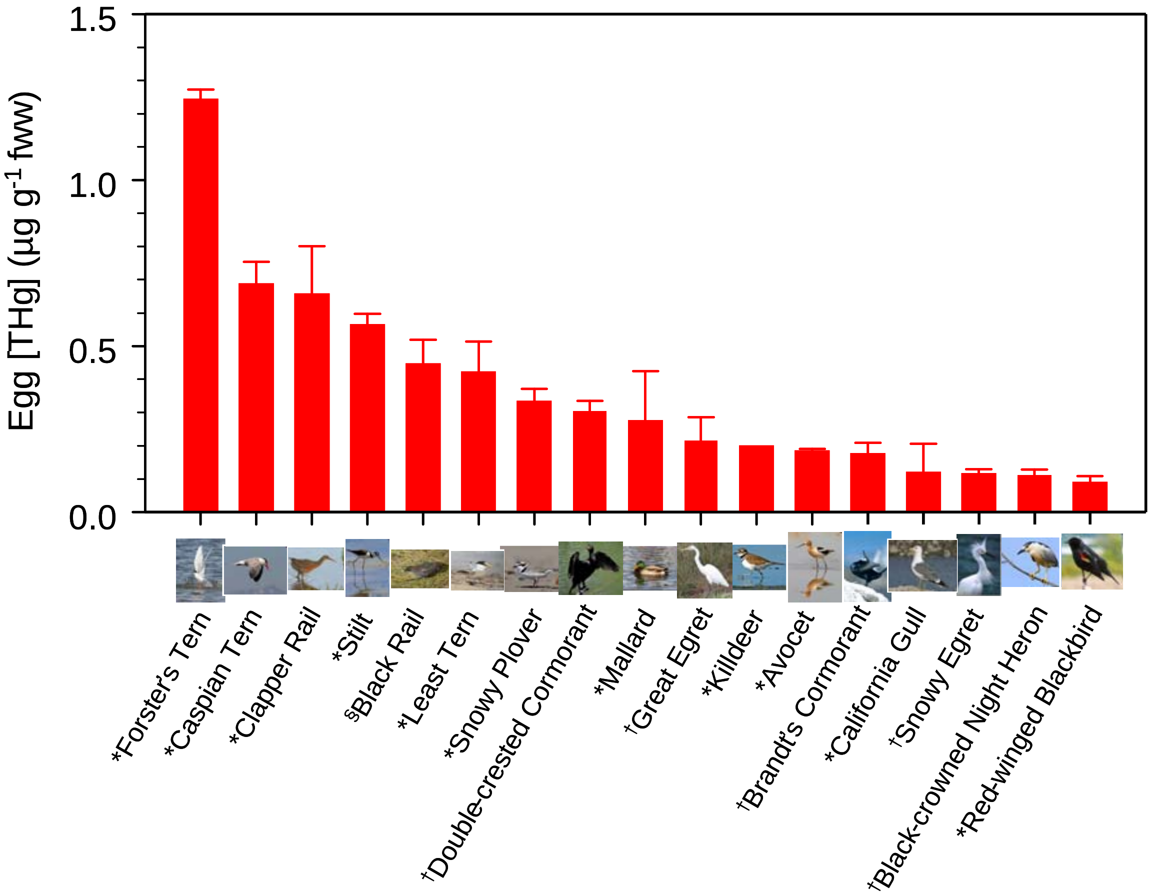


Figure 10. Geometric ± SE mean mercury concentrations in 17 species of aquatic bird eggs (µg g^{-1} fresh wet weight [fww]) in the San Francisco Bay Estuary, California. Of the birds studied, Forster's Terns have the highest egg mercury concentrations, other fish and invertebrate eating waterbirds have moderate mercury concentrations, and aquatic dependent songbirds have the lowest mercury concentrations. *unpublished data from Ackerman and Eagles-Smith 2008b. †data from Schwarzbach and Adelsbach 2003. §data from Tsao et al. 2008.

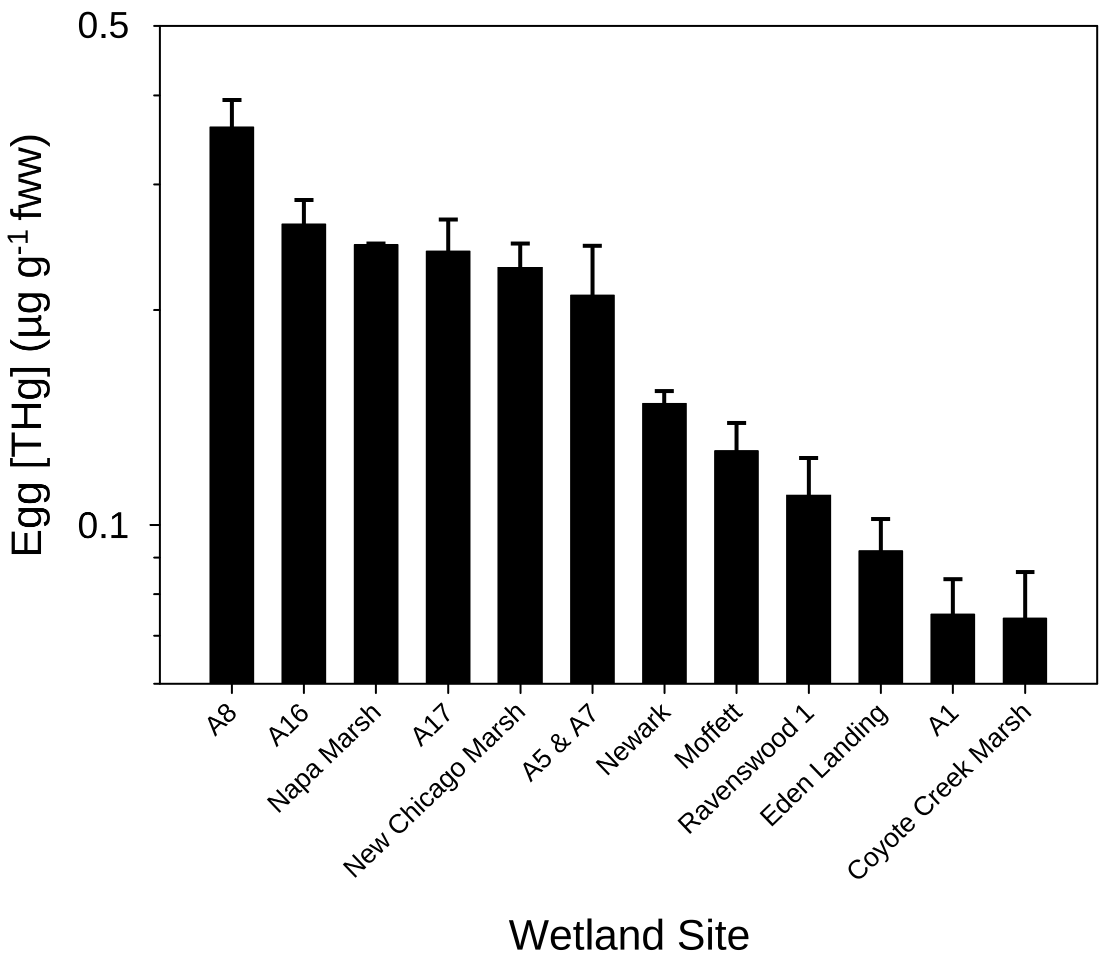


Figure 11. Average±SE mercury concentrations in Avocet eggs collected in San Francisco Bay between 2005-2007. Concentrations vary by up to 5-fold among individual wetlands, highlighting the utility of Avocet eggs to determine wildlife exposure on a pond-specific basis.

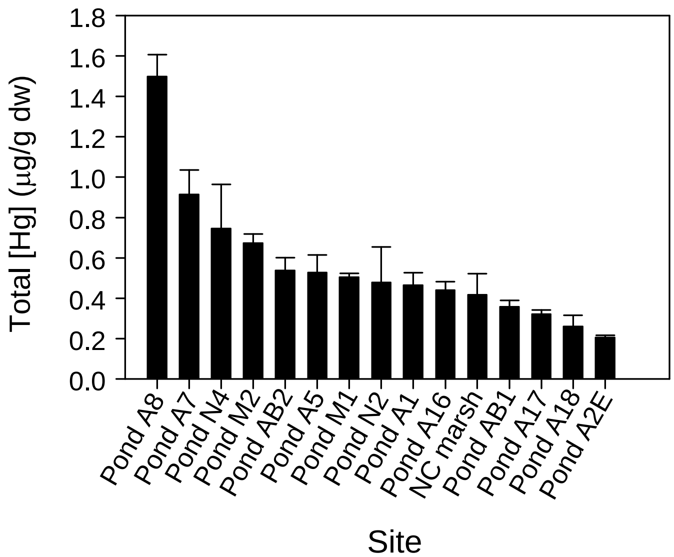


Figure 12. Average (SE) mercury concentrations in three-spined stickleback sampled in saltponds throughout the South Bay Restoration Project Area between 2005-2007.

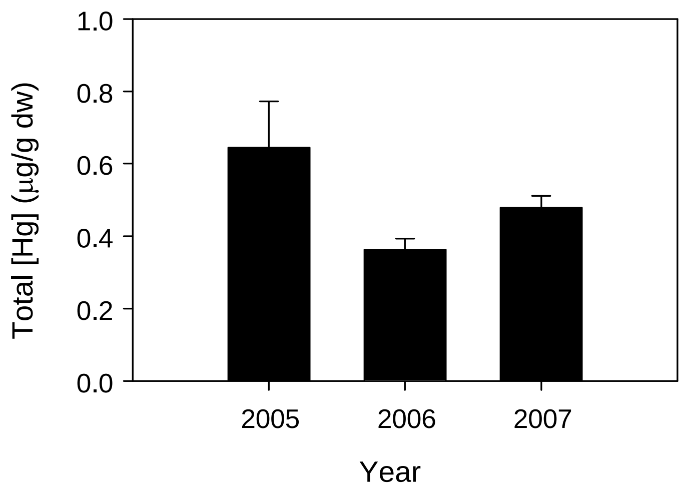
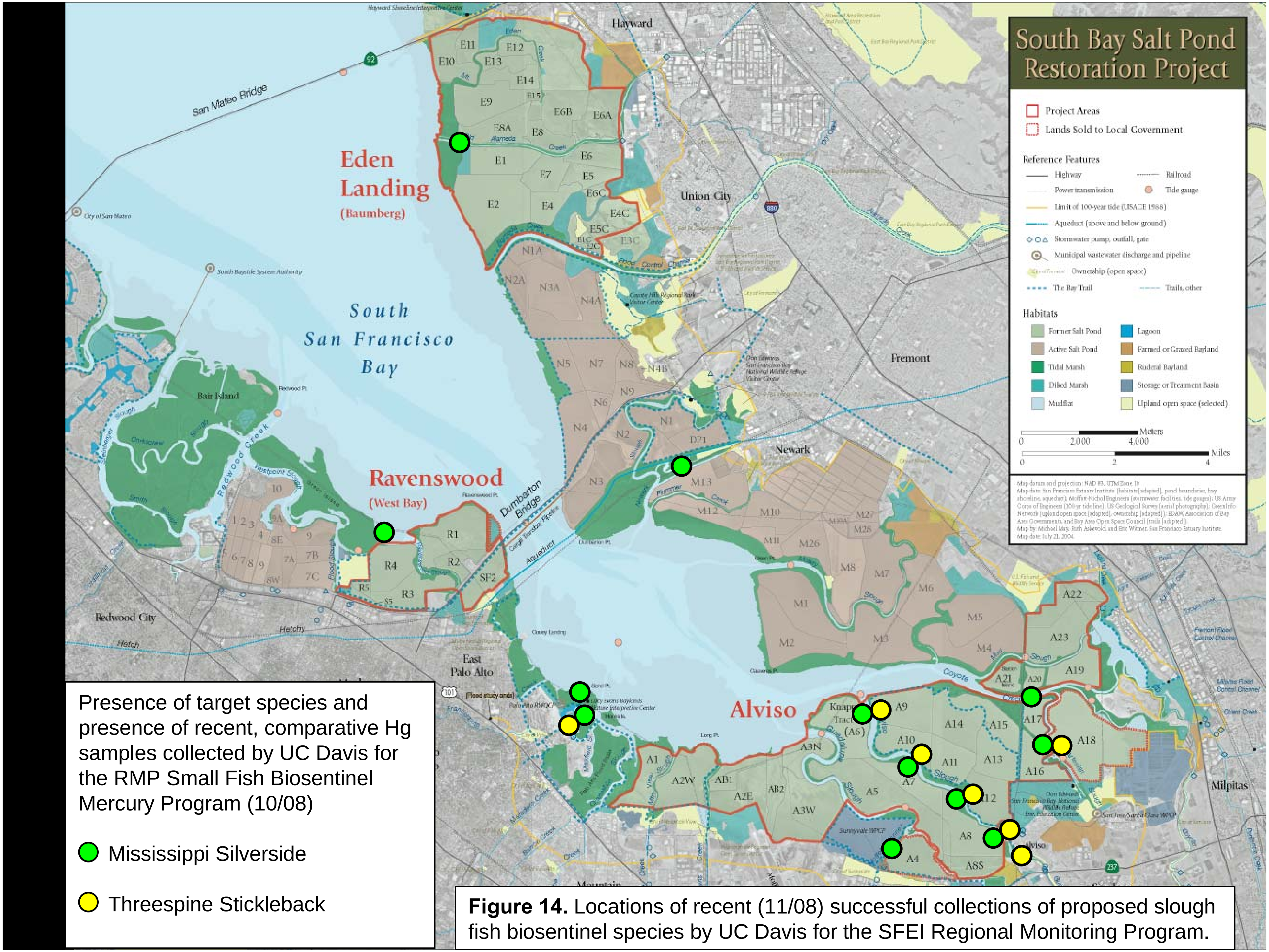


Figure 13. Yearly differences in average (SE) mercury concentrations in three-spined stickleback sampled in saltponds throughout the South Bay Restoration Project Area between 2005-2007.



Presence of target species and presence of recent, comparative Hg samples collected by UC Davis for the RMP Small Fish Biosentinel Mercury Program (10/08)

- Mississippi Silverside
- Threespine Stickleback

Figure 14. Locations of recent (11/08) successful collections of proposed slough fish biosentinel species by UC Davis for the SFEI Regional Monitoring Program.

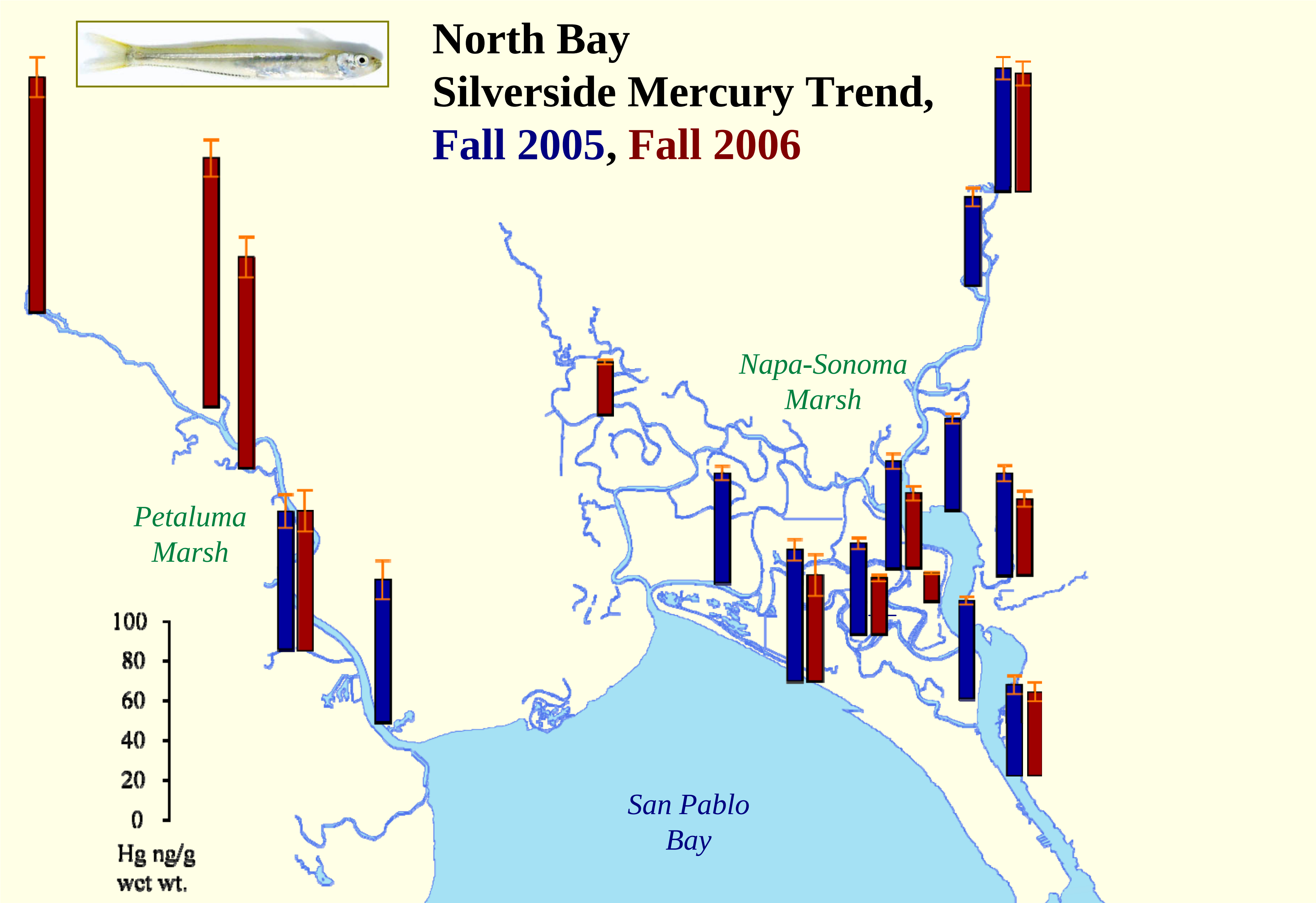


Figure 15. An example of slough fish spatial and inter-annual biosentinel feedback from another region of the Bay (from Slotton et al. 2007). Related work showed this species to also clearly differentiate within-year, seasonal changes in methylmercury exposure, if such changes occur.