

SAN FRANCISCO ESTUARY INSTITUTE

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Re:	Special Studies 2013 – Bioanalytical Tools	
From:	Meg Sedlak and Jay Davis	
To:	Steering Committee	
July 22 nd , 2012		

Recently the State of California convened a national panel to advise the State on Monitoring Strategies for Chemicals of Emerging Concern in Aquatic Ecosystems. The panel recommended that a risk-based system be used to prioritize chemicals to be monitored (see http://ftp.sccwrp.org/pub/download/DOCUMENTS/TechnicalReports/692_CECEcosystemsPanel Report_Final.pdf). Equally important, the panel recognized that evaluations conducted chemical by chemical will not be feasible for monitoring programs. There are approximately 100,000 chemicals in commercial use; monitoring for all of these compounds and developing toxicological profiles for each is cost prohibitive and simply not possible on a time-frame that is relevant to managers. As a result, the panel recommended alternative methods to assess risk through the use of the rapidly growing field of molecular biology. The panel strongly endorsed the development of bioanalytical tools for screening for classes of CECs.

Bioanalytical tools use techniques developed by molecular biologists to identify the presence of compounds that have the potential to adversely affect cellular systems (e.g., hormone signaling, gene replication, etc.). The advantage of such tools is that in one assay, it is possible to screen for the presence of a suite of chemicals that could contribute to a specific toxicological endpoint. Rather than focusing on a chemical-by-chemical basis, bioanalytical tools may make it possible to determine whether contaminants exhibiting a common mode of action (e.g., endocrine disruption) are present in aggregate concentrations that pose concerns. As a result, they are a powerful and potentially inexpensive tool for prioritizing monitoring and management.

The ECWG has requested in the past that the RMP consider bioanalytical tools. Based on this and the state-wide recommendation that bioanalytical tools be developed, SFEI staff solicited a proposal this year from a leader in this field, and member of the State panel, Dr. Nancy Denslow of University of Florida, to develop a bioanalytical tool that is relevant to San Francisco Bay.

The detailed two-year proposal submitted by Dr. Denslow (\$126,000 with \$42,000 match by SCCWRP) is included in the SC package. In brief, this project will link cellular level effects (*in vitro* assays with cell cultures) to whole organism effects (*in vivo* assays) using a species that is relevant to San Francisco Bay, the silverside (*Menidia beryllina*). It is imperative that cellular effects (e.g., up-regulation of a gene) be linked to a higher level effect (e.g. growth, reproduction, or survival of fish) as effects that occur at a cellular level do not assure the occurrence of an adverse effect in an organism. To date, there has been very little work that links responses in bioanalytical tools to responses in marine fish. The expectation is that this research may provide

a critical link that will hasten the development of a bioanalytical tools that can be used for screening effluent and Bay waters.

On its technical merits, the ECWG advisory panel strongly endorsed this proposal. The work is proposed by one of the experts in this field. However, one panel member indicated that while this project is an important step in the process of developing bioanalytical tools for the Bay, it may be a decade before the tool is ready for routine application. Several members of the workgroup expressed concern about this schedule and the fact that it was more of a research project. There was also some question as to why it was compelling to fund it now. Dr. Denslow and SCCWRP currently have approximately \$800,000 to develop bioanalytical tools for evaluating drinking water. It is anticipated that this project would leverage resources and staff from this existing study. There is also strong interest at the State-level to devote resources to the development of these tools.



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July 22nd, 2012

To:Steering CommitteeFrom:Meg Sedlak and David Senn

Re: Special Studies 2013 – Nutrients

As part of the RMP nutrient strategy, the nutrient Scientific Advisory Group (SAG) has reviewed and endorsed a proposal (\$355,000) that includes the following elements:

- Development of a moored sensor monitoring program;
- Development of a Solid Phase Adsorption Toxin Tracking tool for monitoring microcystins and related toxins;
- Continued stormwater nutrient monitoring (augmenting the existing stormwater monitoring effort); and
- Supporting the second year of funding to complete the quantification of nutrient loads into San Francisco Bay.

The TRC endorsed the proposal in concept; however, they had concerns about the funding for the moored sensor element of the proposal. The original proposal submitted to the TRC identified joint funding from the RMP, USGS-Menlo Park, and USGS-Sacramento for the moored sensor study. That task also proposed potential matching funds from other nutrient stakeholders (\$80,000 for hardware purchase). However, those matching funds have not been secured to date, and it is possible that the funding will not be available.

In general, the TRC and SAG feel that it is important to embark on the development of a moored sensor program, and recommended that the full proposal, including the additional \$80,000, be submitted to the SC. The TRC also discussed three potential approaches to funding that the SC may choose to consider if the additional funding is not secured by January 1, 2013:

- Consider requesting \$80,000 from unencumbered reserves to cover the sensors.
- Consider funding this project over a longer time horizon (2 years) to reduce 2013 costs.
- Consider dropping one of the special studies for 2013 to free up additional resources (e.g., the bioanalytical tool project).

It should be noted that the USGS water quality program (Dr. Jim Cloern) had a one-time budget surplus and returned \$35,000 of its 2012 allocation, requesting that these funds be applied to nutrient monitoring program development. If that funding can be applied here, the net request is \$320,000.

TOTAL AVAILABLE FOR SPECIAL STUDIES 2013	\$1,093,540
Total Proposed Studies	\$1,189,000
Remaining Balance	-\$95,460
PROPOSED PILOT AND SPECIAL STUDIES - 2013	
1. EC: PBDE Summary Report	\$35,000
2. EC: Updating RMP Emerging Contaminants Strategy	\$20,000
3. EC: Current Use Pesticide Focus Meeting	\$15,000
4. EC/EE: Linkage of In Vitro Assay Results With In Vivo End Points	\$70,000
5. EE: Developing Benthic Community Condition Indices for Mesohaline Environments	\$76,000
6. EE: Follow up to Moderate Toxicity Workshop (proposal TBD Fall 2012 after workshop - no	
study included in this package)	\$50,000
7. CF: Shared Modeling Proposal	\$100,000
8. STLS: Stormwater Loads Monitoring in Representative Watersheds	\$343,000
9. STLS: Develop and Update Spreadsheet Model - Year 4	\$25,000
10. STLS: Landuse/ Source Area Specific EMC Development	\$80,000
11. STLS: Management Support for Spreadsheet Model Outreach and "Land Use" Based	
Monitoring	\$20,000
12.1 Nutrients: Project Management	\$20,000
12.2 Nutrients: Moored Sensor Monitoring Program Development	\$200,000
12.3 Nutrients: Algal Biotoxin Monitoring	\$65,000
12.4 Nutrients: Stormwater Nutrient Measurements	\$40,000
12.5 Nutrients: Nutrient Loads and Data Gaps	\$30,000

PS/SS: PBDE Summary Report

Estimated Cost:	\$35,000
Oversight Group:	Emerging Contaminant Work Group
Proposed by:	Susan Klosterhaus, SFEI

Background

A PBDE Conceptual Model/Impairment Assessment (CM/IA) report was completed in 2007 to address concerns regarding the bioaccumulation, fate, and potential toxicity of polybrominated diphenyl ethers (PBDEs) in San Francisco Bay (Werme et al. 2007). The report concluded that elevated PBDE concentrations in sport fish and wildlife suggested possible impairment but that a definitive statement was not possible without defined standards (e.g., fish screening values). The report also identified a need for additional PBDE monitoring data in the Bay, particularly information on watershed loading. At that time, only a few years of occurrence data were available.

As a result of incorporating PBDEs into routine monitoring, the RMP now has over ten years worth of occurrence data for San Francisco Bay. New information on the potential for toxicity to Bay wildlife and humans eating contaminated Bay fish is also available. This information includes the release of California sport fish contaminant goals (Klasing and Brodberg 2011) and a bird egg injection study (Rattner et al. 2011), which have largely resulted in decreased concern for PBDE impairment in the Bay. A report that summarizes the current status of PBDE concentrations is needed to communicate to RMP participants, environmental managers, and other researchers that concerns over elevated concentrations in Bay wildlife are being addressed and that available information suggests there is currently no basis for regulatory action.

Study Objective and Applicable RMP Management Question

The objective of this effort is to draft a report that summarizes the current status of PBDEs in San Francisco Bay. This study would address the following RMP management question (MQ):

MQ1. Are chemical concentrations in the Estuary at levels of potential concern and are associated impacts likely?

- A: Which chemicals have the potential to impact humans and aquatic life and should be monitored?
- B: What potential for impacts on humans and aquatic life exists due to contaminants in the Estuary ecosystem?

Approach

SFEI staff will collaborate with the San Francisco Bay Regional Water Quality Control Board to develop the report. The report will include the following elements:

1. Bay occurrence data – The report will include a summary of the following RMP PBDE datasets, including concentration trends over time and congener profiles:

- Surface waters (2002-2011)
- Sediments (2002-2012)
- Deployed bivalves (2002,2003,2005,2006,2008,2010,2012)
- Sport fish (2000,2003,2006,2009)
- Cormorant and tern eggs (2002,2004,2006,2009,2012)

The report will also include the most recent information on the potential for BDE 209 to degrade to more toxic, lower brominated congeners.

2. Relevant toxicity information – The report will compare Bay occurrence data to available toxicity thresholds, including the California sport fish contaminant goals (Klasing and Brodberg 2011), the bird egg injection study funded by the RMP (Rattner et al. 2011), and the Canadian environmental quality goals (Environment Canada 2010), among others.

3. Occurrence data for PBDE replacements – The report will include a short summary of the work to date regarding the occurrence of the PBDE replacement compounds in the Bay.

Budget

Data formatting and analysis	\$15,000
Draft report	\$15,000
Final report	\$5,000
Total	\$35,000

References

Environment Canada. 2010. Risk management strategy for polybrominated diphenyl ethers (PBDEs). Chemicals Sectors Directorate, Environmental Stewardship Branch. Final revised: August 2010.

Klasing, S. and Brodberg, R. 2011. Development of fish contaminant goals and advisory tissue levels for common contaminants in California sport fish: polybrominated diphenyl ethers (PBDEs), Pesticide and Environmental Toxicology Branch Office of Environmental Health Hazard Assessment California Environmental Protection Agency.

Rattner, B., Lazarus, RS, Heinz, GH, Karouna-Renier, NK, Hale RC. 2011. Apparent Tolerance of Common Tern (Sterna hirundo) Embryos to a Pentabrominated Diphenyl Ether Mixture (DE-71). San Francisco Estuary Institute, Final Report. December 2011.

Werme, C., Oram, J, McKee, L, Oros, D, and Connor, M. 2007. PBDEs in San Francisco Bay, Conceptual Model/Impairment Assessment. Prepared for Clean Estuary Partnership. July 31, 2007.

PS/SS: Updating RMP Emerging Contaminants Strategy

Estimated Cost:	\$20,000
Oversight Group:	Emerging Contaminant Work Group
Proposed by:	Susan Klosterhaus, SFEI

Background

The RMP has just completed a synthesis document summarizing the occurrence of contaminants of emerging concern (CECs) in San Francisco Bay (Klosterhaus et al. 2012). In addition to RMP funding, many of the CECs studies to date have been the result of pro bono work conducted as a result of collaborations with universities, government agencies, and commercial laboratories. These opportunities were identified by RMP staff through professional contacts and literature reviews. These studies have allowed for prioritization of these CECs using occurrence and toxicity data to determine the level of concern for individual contaminants in the Bay. The RMP strategy document currently being developed articulates three approaches for identifying CECs for monitoring. These approaches are based on:

- Existing information (known or suspected use, occurrence or toxicity from other locations, best professional judgment),
- Effects (i.e., bioassays), and
- Occurrence (non-target analyses such as the RMP-funded project with NIST or fate modeling).

This will be an iterative process as new information, new analytical methods, and new collaborations become available. In order to keep the CEC Strategy document relevant and timely, funds are needed to review new results, track relevant work being conducted elsewhere, and develop potential collaborations.

Study Objective and Applicable RMP Management Question

The objective of this effort is to insure the RMP is keeping up with the state of the science regarding CECs by tracking new information as it becomes available and communicating relevant information to the ECWG. This study would address the following RMP management question (MQ):

MQ1. Are chemical concentrations in the Estuary at levels of potential concern and are associated impacts likely?

- A: Which chemicals have the potential to impact humans and aquatic life and should be monitored?
- B: What potential for impacts on humans and aquatic life exists due to contaminants in the Estuary ecosystem?

Approach

This effort will involve the review of key information sources throughout the year. These sources include:

- Abstracts of newly published articles in key peer-reviewed journals (e.g., Environmental Science and Technology, Environmental Toxicology and Chemistry, Environment International),
- Documents produced by other programs (e.g., USEPA, Environment Canada, European Chemicals Agency, Great Lakes CEC Program),
- Abstracts and proceedings from relevant conferences (e.g., Society of Environmental Toxicology and Chemistry, International Symposium on Halogenate Persistent Organic Pollutants (Dioxin), International Symposium on Brominated Flame Retardants)

The major outcome of this effort will be to provide updates on relevant information to the ECWG each year. More specifically, this information will be used to:

- Propose updates to the tiered risk-management action framework for San Francisco Bay (Klosterhaus et al. 2012),
- Propose additions or removal of CECs on the 'Unmonitored CEC Candidate List' discussed at the May 2012 ECWG meeting, and
- Propose special studies for monitoring new CECs.

It is anticipated that this special study will be conducted each year to insure the RMP is incorporating the most recent scientific findings regarding the monitoring of CECs in the Bay.

Budget

Information gathering from a variety of sources throughout the year	\$20,000
Total	\$20,000

References

Klosterhaus, S., Yee, D., Sedlak, M, Wong, A. 2012. Contaminants of Emerging Concern in San Francisco Bay: A Summary of Occurrence Data and Identification of Data Gaps. RMP draft report. San Francisco Estuary Institute, Richmond, CA.

PS/SS: Current Use Pesticide Focus Meeting

Estimated Cost:	\$15,000
Oversight Group:	Emerging Contaminant Work Group
Proposed by:	Susan Klosterhaus and Don Yee, SFEI

Background

The RMP has been monitoring organochlorine and organophosphate pesticides in San Francisco Bay surface waters, sediments, and wildlife since the program started in the 1990s. In 2008, pyrethroids were added to Status and Trends sediment monitoring as a result of high volume urban use and the potential for toxicity at low concentrations in the environment. Over the years other CUPs have been monitored in water or sediment, with target compounds largely based on suspected use, analytical method availability, and capability for low-level detection. Several of these compounds have been detected in Bay samples (Klosterhaus et al. 2012).

In the last few years, new information on CUPs has become available but RMP staff have not had the resources to stay updated on these developments. There are CUPs that have not yet been considered for monitoring in the Bay, including a number of 'new' compounds (e.g., fungicides or imidacloprid). There are also some compounds that have been recommended for monitoring in surface waters (the pyrethroids bifenthrin and permethrin) (Anderson et al. 2012), but thus far have only been monitored in Bay sediments. Funding is needed for RMP staff to collect and evaluate the new information generated by other programs and researchers to identify potential CUPs of concern that should be proposed for future monitoring by the RMP.

Study Objective and Applicable RMP Management Question

The objective of this effort is to evaluate existing information on the use, occurrence, fate, and toxicity of CUPs and develop a list of CUPs for possible monitoring in San Francisco Bay in 2013 and 2014.

This study would address the following RMP management question (MQ):

MQ1. Are chemical concentrations in the Estuary at levels of potential concern and are associated impacts likely?

- A: Which chemicals have the potential to impact humans and aquatic life and should be monitored?
- B: What potential for impacts on humans and aquatic life exists due to contaminants in the Estuary ecosystem?

Approach

This effort will involve the following steps:

- Gather and review information from key people and organizations already tracking information on the use, occurrence, fate, and toxicity of CUPs in the Bay Area and Delta. These key individuals will include Jan O'Hara (San Francisco Bay Regional Board), Stephanie Fong (Central Valley Regional Board), Kelly Moran (TDC Environmental), Susan Kegley (Pesticide Research Institute), Mike Johnson (AQUA Science), and Don Weston (UC Berkeley). This task will include contacting analytical laboratories to determine CUP method capabilities.
- 2. Convene a focus meeting with key individuals in early 2013 to identify a list of CUPs for future monitoring in the Bay. The desired outcome of this meeting is a proposed list of CUPs for monitoring in Bay surface waters and sediments, including the target matrix. This task will include meeting preparation and distribution of the meeting minutes in lieu of a report.

Budget

Information gathering	\$10,000
Focus meeting	\$5,000
Tota	l \$15,000

References

Anderson, P., N. Denslow, J.E. Drewes, A. Olivieri, D. Schlenk, S. G.I. and S.A. Snyder. 2012. Monitoring Strategies for Chemicals of Emerging Concern (CECs) in California's Aquatic Ecosystems. Costa Mesa, CA.

Klosterhaus, S., Yee, D., Sedlak, M, Wong, A. 2012. Contaminants of Emerging Concern in San Francisco Bay: A Summary of Occurrence Data and Identification of Data Gaps. RMP draft report. San Francisco Estuary Institute, Richmond, CA.

PS/SS: Linkage of In Vitro Assay Results With In Vivo End Points

Estimated Cost: \$70,000 for 2013 (Year One). This is proposed as a two-year study; \$56,000 will be requested for 2014, pending acceptable progress in Year One.

Oversight Group:Emerging Contaminants Workgroup and Exposure and Effects Workgroup**Proposed by:**Nancy Denslow (University of Florida) and Keith Maruya and Steve Bay (SCCWRP)

Propose	d Deliverables and Time Line	
Delivera	ble	Completion Date
Task 1:	Convene focus group and develop actionable plan	CSD + 1 month
Task 2:	Develop molecular biomarkers for Menidia	CSD + 4 months
Task 3:	Laboratory tests: Early life stage exposures and in vitro bioassays	CSD + 9 months
Task 4:	Field-collected sample exposures	CSD + 18 months
Task 5:	Chemical analysis of CECs	CSD + 21 months
Task 6:	Reporting	Mid-term (Year 1): CSD + 12 months Final: CSD + 24 months

BACKGROUND

A growing number of contaminants of emerging concern (CECs) are found routinely in permitted discharges and their receiving waters. For the few CECs for which analytical methods exist, these methods are still largely in development and only some are routinely performed by commercial services laboratories. As the development and manufacture of chemicals presents an ever changing landscape, those CECs that are produced in high volumes and/or that are capable of being discharged via treated municipal or industrial wastewater effluent or stormwater runoff represent a moving target for environmental quality managers tasked with assessing and/or mitigating their potential for impact.

The CECs of most concern are those which may be potent at trace concentrations (parts per trillion range) and work as endocrine disruptors. Their presence in waterbodies may be harmful to aquatic biota inhabiting these locations. Such endocrine disrupting chemicals can interact directly with soluble hormone receptors or can interfere with the natural synthesis or metabolism of endogenous hormones and thereby impede normal function of these processes in exposed organisms. Most attention has been focused on chemicals which act as estrogens or androgens or their antagonists. Estrogens are important in brain development and programming of tissue differentiation at early time points during development (Feist and Schreck 1996; Lassiter and Linney 2007; Mandiki et al. 2005; Remage-Healey and Bass 2007; Tomy et al. 2009; Vetillard et al. 2006; Zhang et al. 2008).

In our own work, exposure of fathead minnows to concentrations of ethinylestradiol (EE2) at 2 ng/L induced pericardial/yolk sac edema (Johns et al. 2009). The estrogenic mycotoxin zearalenone (exposure range of 2-50 ng/L) also resulted in myocardial edema (Johns et al. 2009). In addition, we analyzed a limited set of gene expression changes including Vtg, which was up-regulated by the two estrogens, steroidogenic acute regulatory (StAR) protein, insulin-like growth factor 1 (IGF-1) and growth hormone (GH) which were also altered. Thus, these genes in target fish species would be viewed as critically important to include in future studies of responses to estrogenic CECs at the molecular level.

Concurrently, novel in vitro methods based on receptor binding or transactivation have been developed that are extremely sensitive to target chemicals acting with the same mode of action, including the potent endocrine disrupting CECs described above. Work is being performed to adapt these in vitro bioassays for water quality assessment and monitoring purposes. Few studies, however, link results from such *in vitro* assays with higher order *in vivo* effects which result in adversity for survival, growth, reproduction, or susceptibility to disease.

The goal of this project is to establish quantitative linkages between the *in vitro* receptor-based assays and traditional endpoints of adversity in a sensitive estuarine fish model, the common silverside (*Menidia beryllina*) which is an established EPA model for estuarine toxicity. As a demonstration, we will focus on estrogenic responses of selected chemicals of interest first in lab exposures (Year 1) followed by exposure to field-collected wastewater treatment plan (WWTP) effluent and estuarine and marine receiving waters (Year 2).

Study Objective and Applicable RMP Management Question

There is considerable concern about the presence of CECs in treated domestic wastewater that is released into the environment and in waters that are reused for irrigation. In California, many WWTPs discharge their treated waters directly into estuaries or into rivers upstream of estuaries. While there are regulations in place for monitoring specific CECs and other regulated chemicals, there is currently not much being done to discover new CECs in water or assess whether the presence of the CECs may adversely affect aquatic biota. If these experiments are successful, we should be able to start to develop methods to integrate results from *in vitro* assays into monitoring programs and determine how they relate to *in vivo* adverse effects. Results from this study will begin to enable managers to determine whether or not additional action is necessary for treated effluents that are discharged into sensitive estuarine environments. This work will not only be important for California, but also for other states that discharge domestic wastewater into marine/estuarine environments.

The objective of this effort is to develop a tool that will assist in the identification of chemicals of emerging concern that are adversely affecting biota. This study would address the following RMP management question (MQ):

MQ1. Are chemical concentrations in the Estuary at levels of potential concern and are associated impacts likely?

A: Which chemicals have the potential to impact humans and aquatic life and should be monitored?

B: What potential for impacts on humans and aquatic life exists due to contaminants in the Estuary ecosystem?

Study Plan

This study will test estrogenic chemicals that were recently recommended for monitoring in California's receiving waters by the State's Science Advisory Panel for CECs (Anderson et al. 2012), e.g. estrone (E1), bisphenol A (BPA), 4-nonylphenol (4-NP) and galaxolide (HHCB). Traditional *in vivo* endpoints for early life stages of silversides (*M. beryllina*) will include: development, growth, and survival and for juveniles: growth, survival and biochemical endpoints such as plasma vitellogenin and hormone concentrations (Vtg, E2 and T) and hepatic gene expression for at least 5 genes per life stage. We will index estrogen equivalency concentrations required for altering higher order endpoints with biochemical responses within the fish and responses obtained with commercially available estrogen receptor (ER) transactivation assays (see also Task 3). These linkages will enable the use of *in vitro* assays as measures

of both exposure and effect. The concentrations required for both *in vivo* and *in vitro* assays will be quantified to determine reference concentrations above which effects may be expected.

Tasks 1 through 3 will be completed in Year One with 2013 funds. Pending approval from the TRC and ECWG/EEWG, Tasks 4 through 6 will be completed in Year Two with 2014 funds.

Task 1: Initial meeting for project coordination. The study will be initiated by a meeting between the two laboratories to discuss the details of the experimental approach to be used and to set up the time frame for the various experiments.

Task 2: Development of molecular biomarkers for *Menidia beryllina*. There are a few molecular biomarkers already developed for *M. beryllina* including Vtg, ER alpha (esr1), ER beta a (esr3), and androgen receptor (AR), among others (Brander 2011). We will validate these assays with our own samples and also develop additional molecular assays for the following genes: IGF-1; StAR; GH; brain aromatase (cyp19b); and two genes involved in testis differentiation, anti-Mullerian hormone (amh) and doublesex and mab-3 related transcription factor 1 (dmrt1). We will get sequences for these additional genes from a high throughput DNA sequencing experiment, discussed in advance with Drs. Brander and Connon, experts involved in developing a transcriptome for Menidia. We will make all sequences we obtain accessible to these colleagues for their independent studies. The assays that we will develop will be based on quantitative reverse transcriptase polymerase chain reaction (Q-PCR) and involve the quantitative amplification of specific genes from total RNA extracted from early life stage fish or juveniles. These genes have been determined in studies of other fish to be responsive to estrogens *in vivo* (Filby et al. 2007; Ijiri et al. 2008; Johns et al. 2009; Kobayashi et al. 2003). The assays will be validated with standard curves after determining that amplification is proportional to amount of transcript present in the original sample.

Task 3: Laboratory exposures and in vitro bioassays. These experiments will be performed at the University of Florida and SCCWRP. We will divide up the exposures and each will perform a positive control. Exposures will be in the water with at least 5 concentrations of each chemical and a single concentration of 5 ng EE2/L as the positive control. The chemicals that will be tested include E1, 4-NP, BPA, and galaxolide (HHCB). Since Menidia are a sensitive species we will first use a range finding experiment to determine the LC50 and conduct our exposures at or below that level. We will then include two concentrations below and two above this value. The test chemicals will be mixed with a small volume of triethylene glycol (TEG) as a carrier to ensure the chemical gets into the water phase. Dilution water will dechlorinated tap water adjusted to 15 ppt salt (using Instant Ocean) and temperature will be controlled to 20 °C, following standardized test guidelines for early life testing (EPA, 1995).

Each lab will do two types of exposures: early life stages, where we will buy recently hatched embryos and allow them to grow under the chemical conditions described above. For these assays we will place twenty 10-d old larvae per 200 ml beaker in quadruplicate for each condition, including artificial seawater with and without carrier, the positive control and 5 concentrations of each test chemical. The exposure will extend for 7 days. Fish will be fed newly hatched brine shrimp. End points will be mortality and growth. We will also take at least 5 individuals from each beaker to determine changes in expression for the following genes: IGF-1, CYP19B, GH, dmrt1, amh.

For the Juvenile Test, we will use 50-d old fry and place 10 per 500 ml beaker under the chemical and replicative conditions described above. This period will be just before gonadal differentiation in this species, another time of vulnerability to endocrine disruptors. The fish will be exposed for 10 days in a static renewal system in 15 ppt salinity. Fish will be fed tetramin flakes and supplemented with brine shrimp. The endpoints for this assay will include measurements of whole body homogenate

concentrations of Vtg, E2 and T (if the hormone evaluations are possible at this age) and hepatic measurements of gene expression for Vtg, ER α , ER β , AR, and StAR using Q-PCR. We will also perform histopathology on the gonads to distinguish males from females and if we can find a sequence for dmrt1, we will correlate its expression with sex of the fish. In some species this gene marks genetically determined sex (Nanda et al., 2002).

For the *in vitro* assays, we will use the same final concentrations as described above, except that the chemicals will be added to the culture as 1/10 volume of a 10X solution, in order not to dilute out the nutrients required for the tissue culture. We will use the same commercial assays for estrogen receptor (ER) transactivation that will be used in a project recently funded by CA (K. Maruya, PI and N. Denslow, CoI, among others) entitled "Evaluating bioanalytical methods as screening tools for monitoring of CECs in California recycled water applications." There will be significant savings in leveraging the funded project as all assay development and validation for the in vitro assays will occur in that project. We will simply apply the assay in this one.

Expectations and Alternative Strategies. We expect that the concentrations of model compounds that we have selected will show higher order effects in survival and growth in the *in vivo* tests (at least at the higher concentrations tested). From past experiments, we are confident that these concentrations will also impact molecular endpoints within the fish resulting in alteration of gene expression. Lastly, the selected concentrations should also be potent enough to change expression of the high throughput assays. *Menidia* are expected to be as sensitive, if not more sensitive that fathead minnows to these chemicals. In the event that they have high mortality in the selected concentrations, we will repeat the experiment at lower concentrations.

Task 4: Exposure to WWTP effluent and receiving water samples and in vitro bioassays

In Year 2, we will test samples of WWTP effluent and receiving waters from two sites, one in southern California and the other in the San Francisco Bay estuary. A sufficiently large volume of treated final effluent and receiving water from each site will be filtered through a sorbing phase (e.g. C18 or Oasis HLB cartridge) to capture organic contaminants and subsequently eluted by organic solvents. A portion of each eluent will be set aside for analytical chemistry and the remainder will be shipped to one or the other of the two participating laboratories where the eluent will be reconstituted to the same proportional volume as the original sample and tested with either an early life stage assay or a juvenile assay.

One sorbing cartridge each will be processed and shipped to SCCWRP and the University of Florida. The cartridges will be eluted and then air dried in order to reconstitute test solutions to 1X, 5X and 10X the concentrations equivalent to what they were at the field site. Each solution will be tested in triplicate and we will use the 5 ng EE2/L as a positive control. We will perform early life stages and juvenile tests as described above. We will also perform in vitro nuclear receptor transactivation assays as described above, but in this case using the concentrates from the field locations.

Expectations and Alternative Strategies.

Based on the results from Task 2 in year 1, we may adjust the concentrations of the positive controls and we may adjust the concentrations of the reconstituted test solutions. If we find that Menidia are more sensitive to the chemicals than we anticipated, all concentrations will be diluted by 10 to get in the range where we do not see more than 10% mortality. Under these conditions, we expect that all endpoints will be viable and that we will be able to compare estrogenic equivalencies across experiments. We may not see much with field waters for androgenic changes, as most of the CECs that target this axis react as anti-androgenic chemicals.

Task 5: Chemical analysis of CECs

Estrone (E1) and EE2 will be measured by ELISA following the methods of Huang and Sedlak (2001). Galaxolide (HHCB), bpA and 4-NP will be measured by GC-MS after extraction and derivatization as described in Ligon et al. (2008). Samples of sufficiently large volume will be collected to ensure the appropriate sensitivity of measurement, based on the range of treatments (lab) and expected receiving water concentrations. An equivalent amount of chemical as evaluated for the in vivo assays will also be assessed for the in vitro assay.

Task 6: Reporting

We plan to submit a mid-term progress report at the end of year 1 and a final report at the end of year 2. We expect that we will be able to derive relationships between the different levels of results, from the molecular high throughput assays to in vivo molecular endpoints and to in vivo higher order changes in survival, growth and development. The estrogenic and androgenic equivalencies that we will derive will help establish the usefulness of high throughput assays as a means to test the quality of estuarine water. We expect this demonstration project to show the usefulness of the approach.

Projected roadmap from this special study to implementation of in vitro assays in monitoring.

- July 2012 Development of protocols for commercially available bioanalytical tools. (sponsored by the State Water Board). Currently there are at least 3 commercial companies that have developed assays for nuclear hormone receptors, including InVitrogen, Biodetection Systems, and SwitchGear. We are in the process of testing the assays and will choose one system for further work. This is part of a project awarded to SCCWRP by the State Water Board (SWB). Initial studies with all three assays indicate that they work well. We will choose a method that has the best chance for implementation by end users in California.
- 2. Dec 2012 Testing of bioanalytical assays for multiple molecular pathways with concentrates extracted from water. (sponsored by State Water Board). The assays include molecular pathways that are known to be affected by exposures to estrogens, androgens, progesterone and glucocorticoids, among other chemicals. We will use the selected assays to examine extracts of WWTP effluents and compare the results with analytical chemistry of the same extracts. We also plan to participate in a parallel project to use bioanalytical techniques to measure hormone activities in Australian WWTP effluent and surface waters (funded by WERF).
- 3. **April 2013 Implement Menidia bioassays following EPA protocols.** (sponsored by RMP) Tasks 1-3 of proposed project. Determine concentrations of E1, 4-NP, BPA, and galaxolide (HHCB) that show effects *in vivo*. Compare these concentrations to those required to change expression of genes *in vivo* and with concentrations required to activate *in vitro* assays showing ER activity.
- 4. July 2013 Start Bight '13 special study utilizing in vitro assays in conjunction with other monitoring tools. (sponsor TBD). Conduct study using southern California samples that is complementary to concurrent RMP study.
- 5. April 2014 Compare in vivo and in vitro assays for effluent and receiving water. (sponsored by RMP). Complete tasks 4-5 of proposed project.
- 6. September 2014—Integrate results of the proposed RMP *Menidia* project with results from other projects. (co-sponsored by RMP, SCCWRP and partners, SWB, WERF) This would include other endpoints covered in the other funded studies, e.g. those that are planned in conjunction with Bight 2013, future RMP surveys, and SWAMP special studies.
- 7. October 2014 Workshop to teach bioanalytical methodology. (sponsor TBD) Transfer of *in vitro* assay technology to public and private laboratories. This would include entities interested in applying the approach in monitoring programs.

- 8. January 2015 Start interlaboratory study to compare bioanalytical methods to current chemical analysis and *in vivo* tests. (sponsor TBD).
- 9. March2016 Report on interlaboratory study. If performance is deemed satisfactory, develop guidance to implement bioassays in routine monitoring.
- 10. July 2016 Start pilot implementation study. Identify partner agencies and facilitate incorporation of in vitro assays into their monitoring programs on a limited basis.
- 11. July 2017 Assess pilot study results. Review results from pilot study and develop plans and guidance for implementation of assay methods.

Potential partners, funding agencies, or possibilities to leverage funds for future progression of bioanalytical tool development.

- 1. WERF—applied research
- 2. NOAA using Mussel Watch as an environmental effects platform
- 3. NSF funding of basic research oriented projects
- 4. State Water Board future phases for development and implementation of bioanalytical screening tools
- 5. Other Water Quality Stakeholders special studies as part of regional surveys (e.g. Bight 2013; RMP) and specific discharger needs (e.g. outfall maintenance)

Budget

The scope of this study will require two years, with Year 1 devoted to obtaining molecular biomarkers for *Menidia* and defining critical endpoints of adversity in early life stage fish. Year 2 will focus on application of these endpoints to effluent and receiving water samples (see Deliverables and Time Line). We are requesting a total budget of \$126,000, with \$70,000 for work to be completed in 2012 and the remaining \$56,000 for 2013. In addition, SCCWRP will contribute leveraged funds via two projects, one supporting the development of in vitro bioassays (or "transactivation assays") funded by the State Water Board and scheduled for completion in mid-2013, and from an internally funded project to develop in vivo bioassays using Menidia and other estuarine marine fish species.

Project Budget

Description	Cost per Sample (\$)	Total Cost Estimate (\$)	Cost Estimate minus match (\$)
Yr: 1			
Development of Molecular Biomarkers		22,000	22,000
Early life stage assay—model compounds 4 (SCCWRP Match – 8,000)	6,000	24,000	16,000
Juvenile assay – model compounds 4 (SCCWRP Match – 8,000)	6,000	24,000	16,000
Transactivation Assays 4	3,000	12,000	12,000
Analytical chemistry (SCCWRP Match 4,000)	2,000	8,000	4,000
Total Yr 1		90,000	70,000
Yr:2			
Water extracts (\$6,000 in kind SCCWRP)	6,000	6,000	0
Early life stage assay – field sites – 4 (SCCWRP Match—8,000)	6,000	24,000	16,000
Juvenile assay – field sites 4 (SCCWRP Match 8,000)	6,000	24,000	16,000
Transactivation assays – 4	3,000	12,000	12,000
Analytical Chemistry – Field sites –4	3,000	12,000	12,000
Total yr 2		78,000	56,000
Total Requested Budget		168,000	\$126,000
SCCWRP contribution	\$42,000		

References:

Anderson PA, Denslow ND, Drewes JE, Olivieri AW, Schlenk D, Scott GI, Snyder SA. 2012. Monitoring Strategies for Chemicals of Emerging Concern (CECs) in California's Aquatic Ecosystems: Final Report. SCCWRP Technical Report 692, Costa Mesa, CA.

Brander SM. 2011. From 'Omics to Otoliths: Establishing *Menidia* Species as Bioindicators of Estrogenic and Androgenic Endocrine Disruption. Ph.D. Dissertation, University of California, Davis.

EPA. 1995. Short-term methods for estimating the chronic toxicity of effluents and receiving waters to West Coast marine and estuarine organisms. EPA/600/R-95-136, August 1995.

Feist G, Schreck CB. 1996. Brain-pituitary-gonadal axis during early development and sexual differentiation in the rainbow trout, Oncorhynchus mykiss. Gen Comp Endocrinol 102:394-409.

- Huang CH, Sedlak DL. 2001. Analysis of estrogenic hormones in municipal wastewater effluent and surface water using enzyme-linked immunosorbent assay and gas chromatography/tandem mass spectrometry. Environ Toxicol Chem 20:133-139.
- Ijiri S, Kaneko H, Kobayashi T, Wang DS, Sakai F, Paul-Prasanth B, Nakamura M, Nagahama Y. 2008. Sexual dimorphic expression of genes in gonads during early differentiation of a teleost fish, the Nile tilapia Oreochromis niloticus. Biol Reprod. 78:333-341.
- Johns SM, Denslow ND, Kane MD, Watanabe KH, Orlando EF, Sepulveda MS. 2009. Effects of estrogens and antiestrogens on gene expression of fathead minnow (Pimephales promelas) early life stages. Environ Toxicol 26:195-206.
- Kobayashi T, Kajiura-Kobayashi H, Nagahama Y. 2003. Induction of XY sex reversal by estrogen involves altered gene expression in a teleost, tilapia. Cytogenet Genome Res. 101:289-294.
- Lassiter CS, Linney E. 2007. Embryonic expression and steroid regulation of brain aromatase cyp19a1b in zebrafish (Danio rerio). Zebrafish 4:49-57.
- Ligon AP, Zuehlke S, Spiteller M. 2008. GC-MS analysis of organic compounds in wastewater and sewage sludge. J Sep Sci. 31:143-150.
- Mandiki SN, Babiak I, Bopopi JM, Leprieur F, Kestemont P. 2005. Effects of sex steroids and their inhibitors on endocrine parameters and gender growth differences in Eurasian perch (Perca fluviatilis) juveniles. Steroids 70: 85-94.
- Nanda I, Kondo M, Hornung U, Asakawa S, Winkler C, Shimizu A, Shan Z, Haaf T, Shimizu N, Shima A, Schmid M, Schartl M. 2002. A duplicated copy of DMRT1 in the sex-determining region of the Y chromosome of the medaka, Oryzias latipes. Proc Natl Acad Sci U S A. 99:11778-11783.
- Remage-Healey L, Bass AH. 2007. Plasticity in brain sexuality is revealed by the rapid actions of steroid hormones. J Neurosci 27:1114-1122.
- Tomy S, Wu GC, Huang HR, Chang CF. 2009. Age-dependent differential expression of genes involved in steroid signalling pathway in the brain of protandrous black porgy, Acanthopagrus schlegeli. Dev Neurobiol 69:299-313.
- Vetillard A, Ferriere F, Jego P, Bailhache T. 2006. Regulation of salmon gonadotrophin-releasing hormone gene expression by sex steroids in rainbow trout brain. J Neuroendocrinol 18:445-453.
- Zhang X, Hecker M, Park JW, Tompsett AR, Newsted J, Nakayama K, Jones PD, Au D, Kong R, Wu RS, Giesy JP. 2008. Real-time PCR array to study effects of chemicals on the Hypothalamic-Pituitary-Gonadal axis of the Japanese medaka. Aquat Toxicol 88:173-182.

PS/SS: Development of Benthic Community Condition Indices for Mesohaline Environments of the San Francisco Bay.

Estimated Total Cost:	\$125,800 (two-year study 2012 and 2013) \$50,000 has been to 2012 activities
Oversight group:	Exposure and Effects workgroup
Proposed by:	Eric Stein SCCWRP

Funding requested for 2013: \$75,800

Introduction and Background

Benthic community assessment is often used as an indicator of ecosystem condition and has become a central element of regulatory programs such as the California's sediment quality objectives for bays and estuaries. Benthos are the indicators of choice for monitoring and assessment for several reasons, including:

- Limited mobility makes them indicative of impacts at the site where they are collected.
- Several animal phyla and classes are sensitive to impacts to their environments and can be used to differentiate certain types of effects.
- Life-histories are short enough that the effects of one-time impacts disappear within a year but long enough to integrate the effects of multiple impacts occurring within seasonal time scales.
- Living in the bottom sediments, benthos have high exposure to common anthropogenic impacts, such as sediment contamination, high sediment organic carbon, and low bottom dissolved oxygen.
- They are important components of aquatic food webs, transferring carbon and nutrients from suspended particulates in the water column to the sediments by filter feeding and serving as forage for bottom-feeding fishes.

For benthic data to be useful in a regulatory context, they must be interpreted in relation to scientifically valid criteria or thresholds that distinguish "healthy" from "unhealthy" benthic communities. While reducing complex biological data to index values has disadvantages, the resulting indices remove much of the subjectivity associated with data interpretation. Such indices also provide a simple means of communicating complex information to managers, tracking trends over time, and correlating benthic responses with stressor data.

To date, benthic indices have been calibrated and validated for two nearshore habitats in California, 1) southern California marine bays, and 2) polyhaline (high salinity) portions of San Francisco Bay. Indices have not been developed for other habitats such as the low salinity mesohaline and tidal freshwater environments. These habitats are particularly challenging because they are naturally subject to relatively broad ranges of conditions (e.g. salinity and dissolved oxygen) and hence the resident organisms are adapted to tolerate environmental stress.

These challenges can be addressed through compilation of robust data sets, careful identification of reference conditions to anchor indices and development of multiple indices that can be used to increase overall sensitivity to detect change in condition.

The objective of this project is to develop and calibrate a minimum of three benthic indices for the mesohaline environments of San Francisco Bay. To the extent possible, we will use the initial consultations with experts to provide a foundation for future work on developing an index for the tidal fresh environment. We do not anticipate that the currently available funds are sufficient for full index development. Therefore, we have divided this proposal into two phases that could be independently funded and would each produce defined products.

Study Objective and Applicable RMP Management Questions:

The objective of this effort is to develop an index for the mesohaline portions of the Bay. This study would assist in our ability to answer the following priority questions for benthos:

- 1. What are the spatial and temporal patterns of impacts of sediment contamination?
- 2. Which pollutants are responsible for observed impacts?
- 3. Are the toxicity tests, benthic community assessment approaches, and the overall SQO assessment framework reliable indicators of impacts?

Overall Study Approach

We will focus on the development of three indices. The first two indices are based on species composition of large numbers of species: the Benthic Response Index (BRI; Smith et al., 2001; Smith et al., 2003; Ranasinghe et al., 2009) and the River Invertebrate Prediction and Classification System (RIVPACs; Wright et al., 1993; Van Sickle et al., 2006; Ranasinghe et al., 2009). The third index will be a multimetric index (MMI) based on community measures and indicator species (e.g. the Index of Biotic Integrity; IBI; Thompson and Lowe, 2004; Ranasinghe et al., 2009).

Index development will be divided between two phases that can be independently funded and that each have independent products.

Phase 1 (to be completed in 2012 with already approved 2012 funding)

Task 1: Update database. Update the existing 2,200 sample 1992 to 2008 standardized taxonomy benthic database with recently collected benthic data and associated habitat (salinity, depth, sediment grain size distribution, and total organic carbon), chemical contaminant, and sediment toxicity data.

Task 2: Refine Habitat Definitions. Refine the San Francisco estuary and delta habitat definition scheme to facilitate the application of SQOs. The original SQO habitat boundary definitions were based on about 140 San Francisco Bay samples in a 714 sample U.S. west coast data set (Ranasinghe et al., In Press). A subsequent study based on more than 501 San Francisco Bay samples (Thompson et al., In Revision) identified potential subtle differences that should be evaluated and defined for SQO implementation. The differences include (1) potential differences in the areal definitions of existing habitats, especially at the Bay margins, and (2) potential addition of an oligohaline habitat between the mesohaline and tidal freshwater habitats in the Suisun Bay region.

Item #7 RMP Study #5Page 3 of 5EE: Developing Benthic Community Condition Indices for Mesohaline Environments

Task 3: Identify and Withhold Validation Data and conduct BPJ study. For each new habitat, identify about 20 samples covering the entire range of habitat conditions for inclusion in a BPJ benthic expert study. These samples will be withheld from the calibration data set in order to assure independence of the two data sets. Experts will be asked to (1) rank the validation samples from least to most disturbed, based on species abundance and minimal habitat data, (2) assign each sample to one of four assessment categories, based on existing narrative definitions, and (3) identify sample characteristics used to rank and categorize the samples.

Budget, Schedule, and Deliverables

The main products of Phase 1 would be the database of organisms and their associated habitats and the results of the BPJ study, which would allow the designation of "good" vs. "bad" locations based on taxonomic composition. At the conclusion of Phase 1, we would also produce a general roadmap for next steps in constructing the benthic indices.

The total cost to complete the Phase 1 tasks would be \$35,323 (Table 1). We anticipate the work would take approximately 3-4 months to complete depending on quality of the data sets and the availability of the expert panel for the BPJ exercise.

Task	Description	Total
1	Update Database	6,894
2	Refine Habitat Definitions	2,219
3	Identify and Withhold Validation Data and conduct BPJ study	26,209
Total		35,323

Table 1. Phase 1 Budget

Phase 2 (to be initiated with 2012 funds; however, bulk of the funding is being requested from 2013 funds)

Task 4: Develop and Calibrate Benthic Indices. Develop and calibrate indices based on relative site conditions using the standard statistical methods appropriate for each index.

Task 5: Assure Independence of Indices and Habitat Factors. Each calibrated benthic index will be tested for independence from habitat variables such as salinity, sediment grain size distribution, sample depth, latitude, longitude, and total organic carbon. While it is generally accepted that current models of benthic response do not discriminate between chemical contamination and other sources of stress (Borja et al., 2003) this approach will ensure that indices are not driven by habitat factors. The goal is an index responsive to benthic community condition, rather than being driven by one or two habitat factors such as sediment grain size distribution or sample depth.

Task 6: Calculate Benthic Index Values. Benthic index values will be calculated for the validation samples, recently acquired data, and new indices for previously acquired data by applying formulae developed during index calibration.

Task 7: Evaluate and Validate Benthic Indices. The BPJ study results and index values calculated by applying formulae developed by index calibration will be used to validate the indices, establish threshold index values for assessment category assignment, and select the most accurate index or combination of indices for application of SQOs.

Task 8: Prepare Summary Report/Journal Article: Results of the study will be written up for publication to a peer-reviewed journal. The journal manuscript will serve as the final project deliverable.

Budget, Schedule, and Deliverables

The main products of Phase 2 would be validated benthic indices for the mesohaline habitat and the journal manuscripts that document their development and testing process.

The total cost to complete the Phase 2 tasks would be \$90,508 (Table 2); \$75,800 is being requested for work in 2013. We anticipate the work would take approximately 6 months to complete following the completion of Phase 1.

	Table 2.	Phase	2 Budget
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Task	Description	Total
4	Develop and Calibrate Benthic Indices	26,354
5	Assure Independence of Indices and Habitat Factors	8,785
6	Calculate Benthic Index Values	8,785
7	Evaluate and Validate Index	20,459
8	Prepare Journal Article/Project Report	26,126
Total		90,508

Literature Cited

- Borja, Á., Franco, J., Perez, V., 2000. A marine biotic index to establish the ecological quality of soft-bottom benthos within european estuarine and coastal environments. Marine Pollution Bulletin 40, 1100-1114.
- Borja, Á., Muxika, I., Franco, J., 2003. The application of a marine biotic index to different impact sources affecting soft-bottom benthic communities along european coasts. Marine Pollution Bulletin 46, 835-845.
- Hunt, J.W., Anderson, B.S., Phillips, B.M., Tjeerdema, R.S., Taberski, K.M., Wilson, C.J., Puckett, H.M., Stephenson, M., Fairey, R., Oakden, J.M., 2001. A large-scale categorization of sites in San Francisco Bay, USA, based on the sediment quality triad, toxicity identification evaluations, and gradient studies. Environmental Toxicology and Chemistry 20, 1252-1265.
- Ranasinghe, J.A., Weisberg, S.B., Smith, R.W., Montagne, D.E., Thompson, B., Oakden, J.M., Huff, D.D., Cadien, D.B., Velarde, R.G., Ritter, K.J., 2009. Calibration and evaluation of

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five indicators of benthic community condition in two California bay and estuary habitats. Marine Pollution Bulletin 59, 5-13.

- Ranasinghe, J.A., Welch, K.I., Slattery, P.N., Montagne, D.E., Huff, D.D., Lee, H., II, Hyland, J.L., Thompson, B., Weisberg, S.B., Oakden, J.M., Cadien, D.B., Velarde, R.G., In Press. Habitat-related benthic macrofaunal assemblages of bays and estuaries of the western United States. Integrated Environmental Assessment and Management.
- Smith, R.W., Bergen, M., Weisberg, S.B., Cadien, D.B., Dalkey, A., Montagne, D.E., Stull, J.K., Velarde, R.G., 2001. Benthic response index for assessing infaunal communities on the southern California mainland shelf. Ecological Applications 11, 1073-1087.
- Smith, R.W., Ranasinghe, J.A., Weisberg, S.B., Montagne, D.E., Cadien, D.B., Mikel, T.K., Velarde, R.G., Dalkey, A., 2003. Extending the southern California Benthic Response Index to assess benthic condition in bays Southern California Coastal Water Research Project, Technical Report 410, Westminster (CA), USA.
- Thompson, B., Lowe, S., 2004. Assessment of macrobenthos response to sediment contamination in the San Francisco Estuary, California, USA. Environmental Toxicology and Chemistry 23, 2178-2187.
- Thompson, B., Ranasinghe, J.A., Lowe, S., Melwani, A.R., Weisberg, S.B., In Revision. Benthic macrofaunal assemblages of the San Francisco Estuary and Delta. Environmental Monitoring and Assessment.
- Van Sickle, J., Huff, D.D., Hawkins, C.P., 2006. Selecting discriminant function models for predicting the expected richness of aquatic macroinvertebrates. Freshwater Biology 51, 359-372.
- Weisberg, S.B., Thompson, B., Ranasinghe, J.A., Montagne, D.E., Cadien, D.B., Dauer, D.M., Diener, D.R., Oliver, J.S., Reish, D.J., Velarde, R.G., Word, J.Q., 2008. The level of agreement among experts applying best professional judgment to assess the condition of benthic infaunal communities. Ecological Indicators 8, 389-394.
- Wright, J.F., Furse, M.T., Armitage, P.D., 1993. RIVPACS: a technique for evaluating the biological water quality of rivers in the UK. European Water Pollution Control 3, 15-25.

PS/SS: Modeling

Estimated Cost:	\$100,000 (requested RMP 2013 funds) \$100,000 (existing RMP 2012 modeling funds) \$200,000 (proposed non RMP Nutrient Strategy funds in
	\$300,000 (proposed non-RMP Nutrient Strategy funds in 2013)
Oversight Group:	Contaminant Fate Work Group and Nutrient Science Advisory Group
Proposed by:	Don Yee and David Senn, SFEI

Background

In joint meetings with members of the Nutrient and Modeling teams on May 1st and June 4th, 2012, modeling needs for different stakeholder efforts (e.g., RMP contaminant fate, RMP nutrient work, Bay Nutrient Strategy), were explored, and some commonalities supporting the use of a shared modeling platform were identified. Although key institutional agreements for the development and maintenance of such shared modeling tools have not yet been reached, a potential path forward is outlined here.

In the May meeting, a recommendation was made to explore adaptation of open source models already used and validated in projects by other agencies in order to minimize the effort and cost of development and to have a partner agency with interest in long-term support of the model platform for the Bay. Delft3D, used by the USGS in modeling sand fate within the Bay and outside the Golden Gate, and used in other areas worldwide for integrated modeling of hydrodynamics, sediment, bioaccumulative contaminants, and nutrients, was identified as a potential tool.

Study Objective and Applicable RMP Management Question

The objective of this effort is to develop models that can be applied to answer questions regarding nutrient and contaminant cycling in the Bay. This study would address the following RMP management questions (MQs):

Nutrients

- 1. Which nutrient sources, pathways, and transformation processes contribute most to concern?
 - a. What is the relative contribution of each loading pathway (WWTP, Delta, non-point source, etc.) to the Bay overall and the Bay's key sub-systems, and how do these loads vary seasonally?
 - b. What is contribution of nutrient regeneration (benthic fluxes) from sediments and denitrification/nitrogen fixation to SF Bay nutrient budgets?
- 2. What nutrient loads can the Bay assimilate (without impairment of beneficial uses)?

3. What future impairment is predicted for nutrients in the Bay?

Modeling/Forecasting

- 1) What patterns of biota exposure to contaminants of concern are forecast for major segments of the Bay under various management scenarios?
- 2) What is the contribution of contaminated Bay margins to Bay impairment?
- 3) What are the projected impacts of Bay margin management actions to Bay recovery?

Approach

Based on discussions with stakeholders, the tasks described here were identified as logical and deliberate steps to developing a sustainable modeling program across a range of contaminants. This approach relies on coordination among multiple initiatives in order to leverage funds. These initiatives include RMP contaminant fate, RMP nutrients, and the Bay Nutrient Strategy. The first several months of the proposed work involves detailed planning to clarify the science needs of important management decisions that will be addressed through modeling, and development of a modeling approach that appropriately targets those needs. This planning period will also allow us to further solidify the coordination between initiatives, and to begin establishing the necessary institutional agreements. In late 2012 or early 2013 model development will commence.

Tasks 1 through 3 will be conducted in 2012 using funds from the previously allocated \$100,000 (2012 funding). Tasks 4 through6 will begin in 2013, funded partly by RMP funds. Tasks 7through 8 are longer term objectives.

Task 1: A technical report will be developed that explores the pros and cons of adopting Delft3D¹ as a model platform. The report will address a range of issues, including:

- a. Thoroughly develop management questions/issues that need to be addressed for contaminants and nutrients and identify the model requirements posed by those management issues. In particular, the question of what output will be needed from a model to address the management questions will be addressed.
- b. Evaluate technical abilities and limitations of Delft3D hydrodynamics, sediment and water quality packages for addressing the management issues.
- c. Estimate cost and time for initially developing the model (calibration, validation); running and maintaining the model; and interpreting scenarios/simulations.

¹ Delft3D is used here as a placeholder; contingent on agreements with a partner agency to develop and maintain as a common platform.

- d. Identify institutional agreements that need to be established for longer-term support of Delft3D as a shared model platform. For example, what is needed to maximize collaboration with USGS and the model developer (Deltares)?
- e. Develop a draft work plan for nutrient and contaminant fate modeling.

Dates: July-October 2012 (includes 1-2 meetings with Modeling Team, and one set of revisions)

Cost: \$35K plus \$10K non RMP funds (includes Dr. Craig Jones' effort and SFEI staff time, and potentially engaging one or two key consultants).

Task 2: Establish a modeling technical team to work with stakeholders to evaluate the work plan laid out in Task 1. This group will provide input on the modeling approach, necessary resolution, parameterization, and calibration/validation for hydrodynamic, sediment, and water quality and contaminant modeling efforts. This team would be utilized across Tasks 2-7.

Date: October 2012 Cost: \$10K

Task 3: Revise white paper and finalize work plan based on workshop input in Task 2. Identify collaborators or consultants, or develop an RFP Date: December 2012 Cost: \$15K

We propose that the remaining portion of the 2012 funds (\$40,000) be combined with proposed 2013 RMP funds (\$100,000), and with matching funds from other efforts (described below), to begin model development in Task 4 in 2012-2013. The ultimate direction taken in Tasks 4-8 will depend on the final approach developed in Tasks 1-3. Thus, the approach below is only broadly described as a proposed path. We propose to update the TRC and SC in Q4 of 2012, and solicit feedback on the suitability of the selected path relative to RMP goals.

Task 4: Develop underlying hydrodynamic & sediment transport model. If explorations in Tasks 1-3 indeed show that Delft3D meets our needs, the existing USGS Delft3D model (grid, boundary conditions) for sand transport might be used as a launch point. A team of collaborators/consultants will be selected to work with stakeholders and SFEI to develop hydrodynamic & sediment transport models. These underlying hydrodynamic and sediment transport models will be the foundation upon which contaminant fate and nutrient/water quality models are developed. An important component of this will include working with water quality and contaminant collaborators on issues related to grid aggregation to adjust the model resolution to levels that are appropriate for the relevant management questions.

Dates: January-June 2013 Approximate Cost: \$130K = \$100K (2012/2013 RMP modeling) + \$30K (non-RMP Nutrient funds) **Task 5:** Develop low-resolution or pseudo-3D nutrient-phytoplankton water quality models for Suisun Bay and South Bay as a test bed for model parameterization. The development of "basic" biogeochemical models has been identified as a high priority project by the nutrient conceptual model technical team to quantitatively synthesize our understanding of the system, test/generate hypotheses, and inform data collection and future modeling and monitoring efforts. Integrated water quality models are often run at lower resolutions than hydrodynamics to allow for sufficiently fast run times to accommodate the calibration of numerous parameters and to allow for analyzing multiple scenarios. Nonetheless they require accurate underlying hydrodynamic inputs. Therefore, grid (and temporal) aggregation will be a critical aspect, requiring coordination between Tasks 4 and 5. Task 5 will use hydrodynamic flows from coarsely aggregated outputs of Task 4. One potential approach is for the nutrient/phytoplankton water quality model to be developed by a consultant in close collaboration with SFEI and the modeling technical team Dates: Model development - January-June 2013

Approximate Cost: \$150K = \$130K (non-RMP Nutrient funds) + \$20K (RMP modeling)

Task 6: Once the model structure is developed, it will be handed off to SFEI staff who will run simulations and further refine the nutrient-phytoplankton model, working with the modeling technical team and the water quality modeling consultant. Work will include: quantitatively synthesizing nutrient load and ambient concentration data (i.e., mass budgets); assessing the relative importance of processes regulating phytoplankton productivity (light limitation, benthic grazing, potential inhibition by NH4, flushing) and nutrient cycling, and performing sensitivity analyses. Parameters with greatest impact will be refined so that model uncertainty is better understood before embarking on more spatially or temporally resolved efforts (Tasks 7 and 8). The experience gained in model development and calibration (e.g., in grid aggregation) can be used to address model uncertainty and applied to later implementation of the model for other contaminants. Dates: Model application and refinement: June 2013-December 2014 Approximate cost 2013: \$150K = \$130K (non RMP Nutrient funds) + \$20K (RMP modeling)

Approximate cost 2014: \$250K (non-RMP Nutrient funds)

Task 7: Develop relatively low-resolution 3D water quality models for particlereactive and bioaccumulative contaminants. An approach analogous to that taken in Tasks 6 will be followed, but a larger share of the focus will be on accurately modeling long-term sediment fate. Because of the long simulation times (decades) necessary to explore the effects of various management actions on contaminant concentrations in sediments and biota, model sensitivity to grid (and temporal) aggregation will also be investigated here so that uncertainty can be characterized. Dates: January 2014-May 2015

Approximate Cost: TBD (~\$50K assemble data, ~\$25K optimize model for sediment fate, \$200K sensitivity testing & scenario runs)

Task 8: Develop 2nd generation nutrient/phytoplankton and contaminant models, and run simulations to evaluate the effectiveness of various management strategies, building on the experience gained in Task 4-7. Dates: 2014-2016 Cost: TBD

STORWATER LOADS MONITORNG IN REPRESENTATIVE WATERSHEDS

Lester McKee, Jennifer Hunt, and Alicia Gilbreath, SFEI, Richmond, CA

ESTIMATED COST:	\$343,000 per year (2013 special and pilot studies budget)
OVERSIGHT GROUP:	Sources Pathways and Loading Work Group (SPLWG)

PROPOSED DELIVERABLES AND TIMELINE

Deliverable	Due Date
Task 1. Project Management (write and manage sub-contracts, track budgets)	Jul-Sep the following year
Task 2. Equipment purchase and prefabrication	Jul-Sep each year
Task 3. Fieldwork	Oct-Apr each year
Task 4. Laboratory analysis	Nov -May each year
Task 5. QAQC / data management	Jun-Sep each year
	(dependent on final
	sampling date)
Task 6. Preliminary data presentation, interpretation, and discussion	Summer/Fall each year

Background

The San Francisco Bay Hg and PCB TMDLs call for a reduction in loads by 50 and 90% respectively. In response, the Municipal Regional Permit for Stormwater (MRP) (SFRWQCB, 2009) (Provision C.8.e.) calls for better quantification of loads of sediments and trace contaminants on a watershed basis and regionally. This is consistent with a long standing recommendation from the SPLWG where six observation watersheds were recommended. selected on the basis of land use and climate (Davis et al. 2000; 2001). This recommendation was reiterated by the SPLWG during 5-year planning (McKee et al., 2008). As such, the Regional Monitoring Program for Water Quality in San Francisco Bay (RMP), through its Sources, Pathways, and Loadings Workgroup (SPLWG), has been conducting tributary loading studies for 10 years beginning WY2003 in Guadalupe River. The focus has been to provide information on sediment and pollutant transport processes and loads in urban watersheds around the Bay (McKee et al., 2004; 2005; 2006a; 2006b; Davis et al. 2007; Oram et al. 2008; McKee et al., 2009; Gilbreath et al., 2012), and for loads coming into the Bay from the Central Vallev via the Sacramento and San Joaquin River Delta (McKee et al., 2001; Leatherbarrow et al., 2005; McKee et al., 2006; David et al. 2009; 2012). Most of the sampling effort has been focused on three SPLWG identified priority locations using a turbidity surrogate methodology recommended by McKee et al. (2001) and McKee et al. (2003): Mallard Island on the Sacramento River; Guadalupe River in San Jose; and the Zone 4 Line A flood control channel in Hayward.

During 2010 the STLS carried out two tasks to support the development of a draft multi-year watershed loading sampling plan (MYP). The first of these tasks "develop criteria and rank watersheds" used GIS to support a statistical classification of watersheds in the Bay Area. Preliminary results provided evidence that there are at least four distinct classes (Greenfield et al., 2010). The second task "Optimize sampling for loads and trends" took advantage of existing temporally resolute (5-15min) data available in Guadalupe River and Z4LA. These data were statistically resampled using a range of sampling designs and loads estimators (mathematical

Item #7 RMP Study #8 STLS: Stormwater Loads Monitoring in Representative Watersheds

formula for loads calculations). The outcomes supported the logical notion that more samples covering a greater number of storms or the use of the turbidity surrogate method provide loads with the greatest accuracy and the least bias (Melwani et al., 2010). At the March 29, 2011 STLS meeting, draft monitoring methods were outlined that included selection of the first four watersheds for monitoring (Sunnyvale East Channel, Guadalupe River, Lower Marsh Creek, San Leandro Creek), turbidity surrogate methods, the use of manual or ISCO sampling design depending on site logistics, 16 samples over 4 storms for 4 years for MRP category 1 pollutants, annual data management, and a report at the end of 3 years). A series of meetings occurred through the spring and summer to discuss and finalize a number of aspects of sampling design including the analyte list (category 1 and category 2 pollutants), discrete versus composite designs for category 1 and category 2, and nuances of site set up at each location. Final design for WY2011 was not completed until October 2011. Final sampling design for WY2012 load monitoring at the four sites was as follows:

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- 1. Turbidity surrogate at all four locations
- 2. Discrete manual sampling for PCBs, Hg, SSC, nutrients, total organic carbon at all four locations, 4 samples per storm for 4 storms plus quality assurance samples (field blanks, field duplicates) and methylmercury, PBDE and PAH at a lower sampling frequency.
- 3. Composite sampling for copper, selenium, carbaryl, fiprinil, pyrethroids, and toxicity at all four locations, 1 sample per storm for 4 storms aiming for 24 sips per sample based on a prediction of storm magnitude plus quality assurance samples (field blanks, field duplicates). For Guadalupe River, given site logistics, composite samples were taken by hand also based on a prediction of storm magnitude
- 4. Continuous stage and flow data were also collected from each location.
- 5. Rain gauges were installed at each sampling location.

During WY2012, due to a late start and a very dry wet season, San Leandro Creek sampling was completed but only 2 out of 4 planned storms were sampled at Lower Marsh Creek and East Sunnyvale Channel, while 3 out of 4 storms were sampled on Guadalupe River. It is proposed that additional samples be added to WY2013 sampling plan so that over a 3 year period, representative samples are taken at each site for a total number of 12 storms sampled. A lessons-learned document, with suggested recommendations for future monitoring, will be developed during the summer 2012.

Objective: This study will implement a small tributaries monitoring in 2 watersheds.

Applicable RMP STLS / MRP Management Questions

Level I RMP, Q3:	What are the sources, pathways, loadings, and processes leading to
	contaminant-related impacts in the Estuary?
Level II RMP, Q3C:	What is the effect of management actions on loads from the most important
	sources, pathways, and processes?
Level III SPL Q2:	What is the watershed-specific and regional total water flow, load of
	sediment, and load contaminants entering the Bay from the urbanized small
	tributaries and non-urban areas draining to the Bay from the nine-county
	Bay Area and are there trends through time?

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Leve's I IV STLS Q1	: Impairment: Which are the "high-leverage" small contribute or potentially contribute most to Bay imp of concern?	
Level IV STLS Q2:	Loads: What are the loads or concentrations of pollu small tributaries to the Bay?	utants of concern from
Level IV STLS Q3:	Trends: How are loads or concentrations of pollutan small tributaries changing on a decadal scale?	nts of concern from
Level IV STLS Q4:	Support management actions: What are the projecter management actions on loads or concentrations of p from the high-leverage small tributaries and where actions be implemented in the region to have the gr	should management

Approach

POC monitoring will be implemented at 2 bottom-of-the-watershed locations in coordination with BASMAA and other project partners according to the Field Manual developed for WY2012 POC monitoring. Essentially, we will follow the WY2012 sampling design with small modifications based on lessons-learned to-date.

Proposed Budget

This projected budget is higher than the \$328,000 available. This budget assumes that we will set up one location and return to a 2nd location already set up in WY2012. This budget assumes monitoring Guadalupe and Richmond pump station. Final budget is dependent on site selection and whether or not we will need to do any set up. For example, if SFEI continues to monitor at Sunnyvale East Channel, or San Leandro, or Guadalupe, no site set up will be necessary.

Deliverable	Estimated cost (spread across 2 watersheds)
Task 1. Project Management (write and manage sub-contracts, track budgets)	\$26,000
Task 2. Equipment purchase and prefabrication	40,000
Task 3. Fieldwork, Site set-up	\$86,000
Task 4. Laboratory analysis	\$130,000
Task 5. QAQC / data management	\$30,000
Task 6. Draft and final report (per MRP requirements)	\$10,000
Shipping and Travel	\$21,000
Total	\$343,000

References

- Davis, J.A., McKee, L.J., Leatherbarrow, J.E., and Daum, T.H., 2000. Contaminant loads from stormwater to coastal waters in the San Francisco Bay region: Comparison to other pathways and recommended approach for future evaluation. San Francisco Estuary Institute, September 2000. 77pp.
- Davis, J.A., Abu Saba, K., and Gunther, A.J. 2001. Technical report of the Sources Pathways and Loadings Workgroup. San Francisco Estuary Institute, September 1999. 55pp.
- Davis, J.A., Hetzel, F., Oram, J.J., and McKee, L.J., 2007. Polychlorinated biphenyls (PCBs) in San Francisco Bay. Environmental Research 105, 67-86.
- David, N., McKee, L.J., Black, F.J., Flegal, A.R., Conaway, C.H., Schoellhamer, D.H., and Ganju, N.K., 2009. Mercury concentrations and loads in a large river system tributary to San Francisco Bay, California, USA. Environmental Science and Technology 28, No. 10, 2091–2100.

Item #7 RMP Study #8

STLS: Stormwater Loads Monitoring in Representative Watersheds

- David, N., Gluchowski, D.C, Leatherbarrow, J.E, Yee, D., and McKee, L.J, 2011. Evaluation of Loads of Mercury, Selenium, PCBs, PAHs, PBDEs, Dioxins, and Organochlorine Pesticides from the Sacramento-San Joaquin River Delta to San Francisco Bay. A Technical Report of the Sources Pathways and Loading Work Group of the Regional Monitoring Program for Water Quality: SFEI Contribution #xxx. San Francisco Estuary Institute, Oakland, CA. xx pp.
- Gilbreath, A., Yee, D., McKee, L., 2012. Concentrations and loads of trace contaminants in a small urban tributary, San Francisco Bay, California. A Technical Report of the Sources Pathways and Loading Work Group of the Regional Monitoring Program for Water Quality: Contribution No. 650. San Francisco Estuary Institute, Richmond, California. 40pp.
- Greenfield, B., Klatt, M., and McKee, L., 2010. Exploratory categorization of watersheds for potential stormwater monitoring in San Francisco Bay. A technical memo for the Sources Pathways and Loading Workgroup of the Regional Monitoring Program for Water Quality. San Francisco Estuary Institute, Oakland, CA.Leatherbarrow, J.E., L.J. McKee, D.H. Schoellhamer, N.K. Ganju, and A.R. Flegal. 2005. Concentrations and loads of organic contaminants and mercury associated with suspended sediment discharged to San Francisco Bay from the Sacramento-San Joaquin River Delta, California RMP Technical Report. SFEI Contribution 405. San Francisco Estuary Institute. Oakland, CA.
- McKee, L., Ganju, N., Schoellhamer, D., Davis, J., Yee, D., Leatherbarrow, J., and Hoenicke, R., 2002. Estimates of suspended sediment flux entering San Francisco Bay from the Sacramento and San Joaquin Delta. Report prepared for the Sources Pathways and Loading Workgroup (SPLWG) of the San Francisco Bay Regional Monitoring Program for Trace Substances (RMP). SFEI contribution 65. San Francisco Estuary Institute, December 2002. 28pp.
- McKee, L., Leatherbarrow, J., Pearce, S., and Davis, J., 2003. A review of urban runoff processes in the Bay Area: Existing knowledge, conceptual models, and monitoring recommendations. A report prepared for the Sources, Pathways and Loading Workgroup of the Regional Monitoring Program for Trace Substances. SFEI Contribution 66. San Francisco Estuary Institute, Oakland, Ca.
- McKee, L.J., Leatherbarrow, J., Eads, R., and Freeman, L., 2004. Concentrations and loads of PCBs, OC pesticides, and mercury associated with suspended sediments in the lower Guadalupe River, San Jose, California. A Technical Report of the Regional Watershed Program: SFEI Contribution #86. San Francisco Estuary Institute, Oakland, CA. 79pp.
- McKee, L., Leatherbarrow, J., and Oram, J., 2005. Concentrations and loads of mercury, PCBs, and OC pesticides in the lower Guadalupe River, San Jose, California: Water Years 2003 and 2004. A Technical Report of the Regional Watershed Program: SFEI Contribution 409. San Francisco Estuary Institute, Oakland, CA. 72pp.
- McKee, L.J., Oram, J., Leatherbarrow, J., Bonnema, A., Heim, W., and Stephenson, M., 2006. Concentrations and loads of mercury, PCBs, and PBDEs in the lower Guadalupe River, San Jose, California: Water Years 2003, 2004, and 2005. A Technical Report of the Regional Watershed Program: SFEI Contribution 424. San Francisco Estuary Institute, Oakland, CA. 47pp + Appendix A and B.
- McKee, L.J., Ganju, N.K., Schoellhamer, D.H., 2006. Estimates of suspended sediment entering San Francisco Bay from the Sacramento and San Joaquin Delta, San Francisco Bay, California. Journal of Hydrology, 323, 335-352.
- McKee, L., Oram, J., Yee, D., Davis, J., and Sedlak, M., 2008. Sources pathways and loadings workgroup: Fiveyear workplan (2008-2012). A Technical Report of the Sources Pathways and Loading Workgroup (SPLWG) of the San Francisco Bay Regional Monitoring Program for Water Quality (RMP): SFEI Contribution #xxx. San Francisco Estuary Institute, Oakland, CA. xxpp.
- McKee, L., Gilbreath, A., and Eads, R., 2009. Concentrations and loads of trace contaminants in the Zone 4 Line A small tributary, Hayward, California: Water Year 2007. A Technical Report of the Sources Pathways and Loading Work Group of the Regional Monitoring Program for Water Quality: SFEI Contribution #563. San Francisco Estuary Institute, Oakland, CA. 57 pp.
- Melwani, A., Lent, M., Greenfield, B., and McKee, L., 2010. Optimizing sampling methods for pollutant loads and trends in San Francisco Bay urban stormwater monitoring. A technical report for the Sources Pathways and Loading Workgroup of the Regional Monitoring Program for Water Quality. San Francisco Estuary Institute, Oakland, CA. Final Draft.
- SFRWQCB, 2009. California Regional Water Quality Control Board San Francisco Bay Region Municipal Regional Stormwater NPDES Permit, Order R2-2009-0074, NPDES Permit No. CAS612008. Adopted October 14, 2009. 279pp.

http://www.waterboards.ca.gov/sanfranciscobay/board_decisions/adopted_orders/2009/R2-2009-0074.pdf

DEVELOP AND UPDATE SPREADSHEET MODEL – YEAR 4

Alicia Gilbreath and Lester McKee, SFEI, Richmond's, CA

ESTIMATED COST:	\$25,000
OVERSIGHT GROUP:	Sources Pathways and Loading Work Group (SPLWG) /
	Small Tributaries Loading Strategy Team (STLS)

PROPOSED DELIVERABLES AND TIMELINE

Deliverable	Due Date
Task 1. Refine the model for inclusion of GIS source layers and EMC	5/1/2013
back calculation data	
Task 2. Revise and complete Mercury and PCB RWSM V2	9/1/2013
Task 3. Model sensitivity analysis	1/1/2014

BACKGROUND

To accurately assess total contaminant loads entering San Francisco Bay, it is necessary to estimate loads from local watersheds. Presently Hg loads entering the Bay from urban stormwater described in the San Francisco Bay TMDL have been estimated by the Water Board by combining BASMAA bed sediment data with now outdated estimates of regional suspended sediment loads. In the case of PCBs, the mass loads in the Bay TMDL were derived from scaling loads from the Guadalupe and Coyote Creek watersheds by area up to the region as a whole. Although these methods were arguably appropriate for planning and TMDL development, the implementation plans of these TMDLs call for improvements of regional scale loads estimates and to assess how these loads might be reduced. These needs are now reflected in the municipal stormwater permit (MRP) (SFRWQCB, 2009) and in the 2nd and 4th questions of the RMP Small Tributaries Loading Strategy (STLS).

"Spreadsheet models" of stormwater quality provide a useful and relatively cheap tool for estimating regional scale watershed loads. These models are based on the simplifying factor that unit area runoff for homogeneous sub-catchments have constant concentrations. Spreadsheet models have advantages over models such as HSPF and SWMM since data for many of the input parameters required by those models do not currently exist, and also require large calibration data sets which take money and time to collect. A spreadsheet model was developed for the Bay Area previously (Davis et al., 2000); however, at that time, there was only local land use specific data on pollutants of concern (POCs) for a drought period during the late 80s and early 90s, and there was no local data on Hg and PCBs. More recently, a spreadsheet model was developed for a watershed in Los Angeles that was able to predict mass emissions to within 8% of measured Zn loads and described options for loads reduction through a focus on "high leverage" areas (Ha and Stenstrom, 2008). Locally Lewicki and McKee (2009) used a combination of methods to make new watershed specific suspended sediment loads estimates, including application of a spreadsheet model for urban areas in which sediment loads were calculated from watershed area and erosion estimates for specific land use classes. In this model, empirical data and regional regression equations were also applied to larger watersheds dominated by non-urban land use. The combination of these

methods produced estimates of sediment loading to the Bay that are presently deemed to be the best. An improved version of this sediment model will be integrated in to the regional watershed spreadsheet model (RWSM) described further below.

During the RMP 2010 calendar year (year 1 of this project), version 1 of the hydrology component of the regional watershed spreadsheet model (RWSM) was developed. Two base hydrology model approaches were investigated: one using runoff coefficients based on land use and the other based on impervious cover. Initial versions of each model were calibrated to local hydrology data from 18 local watersheds with a wide variety of imperviousness, soil, and slope. Recommendations were made to address hydrology model weaknesses. The year 1 report also presented a review of land use and source areas in relation to PCBs, Hg, dioxins, Cu, and Se and provided recommendations for steps to develop event mean concentration (EMC) data to support the input side of the model. The report recommended the model structure for each pollutant, methods to fill data gaps, and priorities (Lent and McKee 2011).

During RMP 2011 calendar year (year 2 of this project), version 2 of the GIS-based hydrology model was developed following Y1 recommendations. In v2, several more calibration watersheds were added to increase the range of watershed characteristics including % imperviousness character. In addition, land use categories were refined and calibration watersheds that included gauge records from time periods that were not congruent with the period of time represented by the land use dataset were removed. The first versions of the Hg and PCB RWSMs were developed using combinations of SoCal EMC data (Hg only) and world soils data (Hg and PCBs) combined with local SSC EMC data (BASMAA, 1995). The Hg load results were consistent with existing estimates at a regional scale but questionable at the scale of individual land uses. For PCBs, the loads were 20x higher than expected on us a regional scale but in the right order relative to our conceptual models for land uses and source areas.

In parallel, the BASMAA Monitoring / Pollutants of Concern (POC) Committee has been discussing and prioritizing work products in relation to the MRP. During 2011, project profiles were developed for addressing MRP provisions c8e.vi (sediment delivery estimate / budget) and c.14 (PBDEs and OC pesticides). Subsequently, BASMAA has asked SFEI to complete work outlined in these project profiles. Since all these tasks are components of what is envisioned to be a single model developed over three years and final report in 2014, this work plan reflects all recommendations and BASMAA work requests in relation to the RWSM that can be accurately budgeted at this time. However, we are careful to explicitly describe products and deliverables in relation to the specific resources allocated by either the RMP or BASMAA.

During calendar year 2012 (year 3 of this project), we are developing a Copper test case model for RWSM. Copper represents a data rich urban contaminant that follows classical source, build-up, and wash off processes in relation to urban land uses in a similar fashion to PAHs and pesticides and parts of the mercury model process. It therefore represents an ideal test case as a step toward model development for other contaminants that are of more interest. There are abundant local land use specific data on copper EMCs

Item #7 RMP Study #9

STLS: Develop and Update Spreadsheet Model- Year 4

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(BASMAA, 1995) and abundant bottom of the watershed calibration data (BASMAA, 1995; RMP loading studies, recent BASMAA/ BACWA studies; other SFEI studies). In addition, there is SPLWG experience and published papers from SoCal (Stenstrom, Stein and coauthors) to aid in the development of the copper model. Additionally in 2012, using RMP funds for the EMC development study, we will develop improved input datasets that will underlie the refinements to the PCB and Hg models of the RWSM (this will be the second version of these models). These datasets will include GIS layers of Hg and PCB sources, as well as EMCs for specific land uses back-calculated from local empirical data and augmented by the world literature where necessary. Integration of these datasets and calibration and verification of the PCB and Hg RWSM version 2 is an effort that will span two years of RWSM funding (the already allocated funds for 2012, and the funds requested in this proposal for 2013), and which will also be supported by the EMC development pilot study (2012 funding). Initial work on version 2 of the Hg and PCB RWSM in 2012 will include reviewing modeling options (more or less land use / source area classes, hybrid sediment/water based models) and preparing a short memo about those options, which will then become a component of the methods section.

Objective:

The overall objective of this 2013 proposed study is to continue to develop and refine mass emissions estimates of Hg and PCBs for the region as a whole draining into the San Francisco Bay using single watersheds for calibration and verification purposes.

APPLICABLE RMP MANAGEMENT QUESTIONS

What are the sources, pathways, loadings, and processes leading to
contaminant-related impacts in the Estuary? What is the effect of management actions on loads from the most
important sources, pathways, and processes?
What is the watershed-specific and regional total water flow, load of sediment, and load contaminants entering the Bay from the
urbanized small tributaries and non-urban areas draining to the Bay from the nine-county Bay Area and are there trends through time?
Impairment: Which are the "high-leverage" small tributaries that contribute or potentially contribute most to Bay impairment by pollutants of concern?
Loads: What are the loads or concentrations of pollutants of concern from small tributaries to the Bay?
Support management actions: What are the projected impacts of management actions on loads or concentrations of pollutants of concern from the high-leverage small tributaries and where should management actions be implemented in the region to have the greatest impact?

Item #7 RMP Study #9 STLS: Develop and Update Spreadsheet Model- Year 4

METHODOLOGY

In 2013, we propose to:

- Refine the RWSM by incorporating spatial data (GIS layers) of PCB and Hg sources (developed with RMP 2012 EMC funding) as input data sets.
- Refine the RWSM by incorporating back calculations of land use-specific EMCs (developed with RMP 2012 EMC funding) as input data sets.
- Revise and complete Hg and PCB RWSM v2 testing and calibration. We will also evaluate model weaknesses through a sensitivity analysis (combinations of more and less source area classes and reasonable ranges of EMCs for each source class, hybrid models) and make any obvious or within budget improvements. Assumption: The model and documentation will not be packaged for external users. Such packaging and creation of supporting documentation (i.e., a user manual) may be a prioritized as a further step.
- Deliverable: 10 page technical memo

There are 2 other project components developed through the STLS that will add value to the RWSM. Pending available funding (non-RMP funding), the tasks are:

- Update the sediment RWSM by developing an erosional rates classification scheme and updating the model with known sediment outputs.
- Develop contaminant profiles and model work plan recommendations for PBDEs and OC pesticides.

Proposed Cost (all labor)		
Task 1	Refine the model for inclusion of GIS source layers and EMC back calculation data	\$5,000
Task 2	Revise and complete Mercury and PCB RWSM V2	\$15,000
Task 3	Model sensitivity analysis	\$5,000
Total for the 4th year		<u>\$25,000</u>
Subsequent years		?

BUDGET (TO BE ADJUSTED AS NEEDED)

REFERENCES

- Davis, J.A., L. McKee, J. Leatherbarrow, and T. Daum. 2000. Contaminant Loads from Stormwater to Coastal Waters in the San Francisco Bay Region: Comparison to Other Pathways and Recommended Approach for Future Evaluation. San Francisco Estuary Institute, Richmond, CA.
- Ha, S.J. and M.K. Stenstrom. 2008. Predictive Modeling of Storm-Water Runoff Quantity and Quality for a Large Urban Watershed. J. Environ. Eng., 134(9), 703-711.
- Lent, M.A. and McKee, L.J., 2011. Development of regional suspended sediment and pollutant load estimates for San Francisco Bay Area tributaries using the regional watershed spreadsheet model (RWSM): Year 1 progress report. A technical report for the Regional Monitoring Program for Water Quality, Small Tributaries Loading Strategy (STLS). Contribution No. 666. San Francisco Estuary Institute, Richmond, CA.
- Lent, M.A., Gilbreath, A.N., and McKee, L.J., 2012. Development of regional suspended sediment and pollutant load estimates for San Francisco Bay Area tributaries using the regional watershed spreadsheet model (RWSM): Year 2 progress report. A technical progress report prepared for the Regional Monitoring Program for Water Quality in San Francisco Bay (RMP), Small Tributaries Loading Strategy (STLS). Contribution No. 667. San Francisco Estuary Institute, Richmond, California.
- Lewicki, M., and McKee, L.J., 2009. Watershed specific and regional scale suspended sediment loads for Bay Area small tributaries. A technical report for the Sources Pathways and Loading Workgroup of the

Regional Monitoring Program for Water Quality: SFEI Contribution #566. San Francisco Estuary Institute, Oakland, CA. 28 pp + Appendices.

Schueler, T., 2003. "Impacts of impervious cover on aquatic systems," *Rep. to Watershed Protection Research Monograph* #1, Center for Watershed Protection, Ellicot City, Md.

SFRWQCB, 2009. California Regional Water Quality Control Board San Francisco Bay Region Municipal Regional Stormwater NPDES Permit, Order R2-2009-0074, NPDES Permit No. CAS612008. Adopted October 14, 2009. 279pp.

http://www.waterboards.ca.gov/sanfranciscobay/board_decisions/adopted_orders/2009/R2-2009-0074.pdf

POC LOADS MONITORING - LANDUSE/SOURCE AREA SPECIFIC EMC DEVELOPMENT Lester McKee, Jennifer Hunt, and Alicia Gilbreath, SFEI, Richmond, CA

ESTIMATED COST: OVERSIGHT GROUP: \$80,000 (2013 special studies budget) Sources Pathways and Loading Work Group (SPLWG) / Small Tributaries Loading Strategy Team (STLS)

PROPOSED DELIVERABLES AND TIMELINE

Deliverable	Due Date	
Task 1: Project Management	Sep 2012 – Dec 2013	
Task 2: Purchase, prefabricate and install field equipment	Oct 2012	
Task 3: Wet weather fieldwork	Nov 2012 – Apr 2013	
Task 4: Laboratory analysis	Dec 2012 –Jun 2013	
Task 5: Data management	Jun-Jul 2013	
Task 6: Reporting	Jul-Oct 2013	

BACKGROUND

The PCB and Hg TMDLs for San Francisco Bay call for improved stormwater loading information and increased application of urban Best Management Practices (BMPs) for reducing pollutant loads and impacts. Since it is impossible to monitor all stormwater inputs to San Francisco Bay (there are more than 250 urban watersheds presently identified), the first report of the SPLWG recommended a combination of monitoring and extrapolation using modeling to develop regional loads estimates (Davis et al., 2001). In addition, Davis et al. identified a need to evaluate the efficacy of local and regional BMPs for influencing stormwater loads trends. These needs are now reflected in the Municipal Regional Stormwater NPDES Permit (MRP) (SFRWQCB, 2009), in the 2009 version of the Small Tributaries Loading Strategy (STLS, 2009), and in the Small Tributaries Loading Strategy Multi-Year Plan (STLS, 2011).

To estimate regional loads, the STLS documents the consensus recommendation to develop a regional watershed spreadsheet model (RWSM) using the methods of Ha and Stenstrom (2008). Data inputs for such a model include rainfall, runoff coefficients, and land use based contaminant event mean concentrations (EMCs). Such empirical monitoring studies have been performed in Southern California by Tiefenthaler et al. (2008) who selected eight representative land use classes based on management needs. They found statistical differences between industrial, recreational, and open space land use classes for suspended sediment, copper, lead, and zinc and no statistical difference between commercial and any category of residential urban land use or transportation.

Unfortunately these Southern California data are not directly applicable to the Bay Area, where PCBs and Hg are the pollutants of highest concern. In the Bay Area, older industrial areas are hypothesized to be more polluted with PCBs than other urban landscapes, whereas for mercury, a broader distribution is hypothesized that includes industrial and commercial areas with higher imperviousness, and older urban areas.

STLS: Landuse/Source Area Specific EMC Development

In 2010 and 2011, the TRC funded the first and second year of development of that modeling platform (Lent and McKee, 2011; Lent et al., 2012). The outcomes of the first year included the development of two parallel hydrological models, one using land use based runoff coefficients and the other using imperviousness based runoff coefficients. The model outcomes were compared to empirical observations in 18 calibration watersheds. Preliminary loads of PCBs, Hg, and sediment were also generated but confidence was low. In 2011, the TRC provided another \$20k to further the development of the model to finalize the hydrological component. In parallel, a literature review was completed as part of the y1 report (Lent and McKee, 2011). Land use and source specific classes were recommended for the regional watershed spreadsheet model (RWSM) structure, existing EMC data from local sources and literature were reviewed and compiled, and methods for land use/ source area specific EMC estimation were proposed. In addition, recommendations were given for improvement of the GIS data shape and line files that will become the basis of the model structure.

During calendar year 2012, the STLS recommended using the \$80k funding for the following uses:

- Develop a Copper test case RWSM,
- Complete GIS layer development for the basis of the PCBs and Hg RWSM,
- Estimate PCB and Hg EMC data for the land use and or source areas developed in the GIS layers,
- Complete and document Version 2 of the PCB and Hg RWSMs,
- Ensure that the STLS EMC spreadsheet model development is developed with strong step wise communication, and with coordination with other BASMAA efforts, in particular the Clean Watersheds for Clean Bay (CW4CB) project and other permit related efforts.

It is anticipated that the outcomes from this year's work will guide the priorities for EMC development in 2013 but following the RWSM master work plan, the options for use of the money are:

- Further refinement of GIS layers,
- Further computations of PCB and Hg EMC data for the land use and or source areas developed in the GIS layers,
- Empirical field data collection of EMC data for specific land uses or source areas

<u>The objective of this study</u> is to generate event mean concentration data for the input side of the Regional Watershed Spreadsheet model.

APPLICABLE RMP MANAGEMENT QUESTIONS

- Level I RMP, Q3: What are the sources, pathways, loadings, and processes leading to contaminant-related impacts in the Estuary?
- Level II RMP, Q3C: What is the effect of management actions on loads from the most important sources, pathways, and processes?
- Level III SPL Q2: What is the watershed-specific and regional total water flow, load of sediment, and load contaminants entering the Bay from the

urbanized small tributaries and non-urban areas draining to the Bay from the nine-county Bay Area and are there trends through time?

- Level IV STLS Q1: Impairment: Which are the "high-leverage" small tributaries that contribute or potentially contribute most to Bay impairment by pollutants of concern?
- Level IV STLS Q2: Loads: What are the loads or concentrations of pollutants of concern from small tributaries to the Bay?
- Level IV STLS Q4: Support management actions: What are the projected impacts of management actions on loads or concentrations of pollutants of concern from the high-leverage small tributaries and where should management actions be implemented in the region to have the greatest impact?

SAMPLING DESIGN / METHODS

Desktop methods

- Step 1. Update as needed the 2012 data base on local and international data on soils and water concentrations in relation to land use and source areas for Hg and PCBs,
- Step 2. Apply further back-calculation methods (recommended from 2012 report), including inverse optimization methods,
- Step 3. Provide regular updates and feedback opportunities to STLS, including discussion of proposed back-calculation methods,
- Step 4 Perform sensitivity analysis, and develop error bars around results (or professional judgment to assign errors or ranges)
- Step 5. Prepare a short (<5 page) summary of methods and results for inclusion in the model documentation

Field methods

- Task 1: Project management
- Task 2: Purchase, prefabricate and install ISCO auto sampling equipment (yet to be determined if triggered by stage or turbidity or a combination) at 2 EMC sites selected in response to the outcomes of the 2011 reconnaissance
- Task 3: Carry out fieldwork during 4 wet season storms at these EMC sites.
- Task 4: Complete laboratory analysis of water samples
- Task 5: Complete data management/quality assurance
- Task 6: Complete interpretative report

BUDGET

\$80,000 (detail to be determined through STLS team meetings)

REFERENCES

- Davis, J.A., Abu Saba, K., and Gunther, A.J. 2001. Technical report of the Sources Pathways and Loadings Workgroup. San Francisco Estuary Institute, September 1999. 55pp.
- Ha, S.J. and M.K. Stenstrom. 2008. Predictive Modeling of Storm-Water Runoff Quantity and Quality for a Large Urban Watershed. J. Environ. Eng., 134(9), 703-11.
- Lent, M., Oram, J., and McKee, L., 2009. Guadalupe Watershed Model: Year 1 Report. RMP Technical Report: SFEI Contribution #564. San Francisco Estuary Institute, Oakland, CA.
- Lent, M.A. and McKee, L.J., 2011. Development of regional suspended sediment and pollutant load estimates for San Francisco Bay Area tributaries using the regional watershed spreadsheet model

STLS: Landuse/Source Area Specific EMC Development

(RWSM): Year 1 progress report. A technical report for the Regional Monitoring Program for Water Quality, Small Tributaries Loading Strategy (STLS). Contribution No. 666. San Francisco Estuary Institute, Richmond, CA.

- Lent, M.A., Gilbreath, A.N., and McKee, L.J., 2012. Development of regional suspended sediment and pollutant load estimates for San Francisco Bay Area tributaries using the regional watershed spreadsheet model (RWSM): Year 2 progress report. A technical progress report prepared for the Regional Monitoring Program for Water Quality in San Francisco Bay (RMP), Small Tributaries Loading Strategy (STLS). Contribution No. 667. San Francisco Estuary Institute, Richmond, California.
- SFRWQCB, 2009. California Regional Water Quality Control Board San Francisco Bay Region Municipal Regional Stormwater NPDES Permit, Order R2-2009-0074, NPDES Permit No. CAS612008. Adopted October 14, 2009. 279pp.

http://www.waterboards.ca.gov/sanfranciscobay/board_decisions/adopted_orders/2009/R2-2009-0074.pdf

STLS, 2011. Small Tributaries Loading Strategy Multi-Year Plan Version 2011. A document developed collaboratively by the Small Tributaries Loading Strategy Work Group of the Regional Monitoring Program for Water Quality in the San Francisco Estuary (RMP): Lester McKee, Alicia Gilbreath, Ben Greenfield, Jennifer Hunt, Michelle Lent, Aroon Melwani (SFEI), Arleen Feng (ACCWP) and Chris Sommers (EOA/SCVURPPP) for BASMAA, and Richard Looker and Tom Mumley (SFRWQCB). Submitted to the Water Board, September 2011 to support compliance with the Municipal Regional Stormwater Permit, provision c.8.e.

(http://www.swrcb.ca.gov/rwqcb2/water_issues/programs/stormwater/MRP/2011_AR/BASMAA/B2_2 010-11_MRP_AR.pdf)

Tiefenthaler, L.L., Stein, E., and Schiff, K., 2008. Watershed and land use-based sources of trace metals in urban stormwater. Environmental Toxicology and Chemistry 27, 277-87.

MANAGEMENT SUPPORT FOR SPREADSHEET MODEL OUT REACH AND "LAND USE" BASED MONITORING

Lester McKee and Jennifer Hunt, SFEI, Richmond, CA

ESTIMATED COST:	\$20,000 (2013 special and pilot studies budget)
OVERSIGHT GROUP:	Small Tributaries Loadings Strategy Team (STLS)

PROPOSED DELIVERABLES AND TIMELINE

	Deliverables	Due date
Task 1	Local STLS meetings	Quarterly 2013
Task 2	Phone conferences for product updates and review	Monthly 2013

BACKGROUND

The RMP Small Tributaries Loading Strategy (STLS) work group provides the framework for planning and coordinating projects for the improvement of pollutant loads information for S.F. Bay. The STLS has met regularly over the last few years to develop and oversee multiple RMP products including the Regional Watershed Spreadsheet Model (RWSM) and the framework for POC long-term monitoring. These elements together provide assurances that the most cost effective information is generated that directly answers our key loading questions.

In 2011, STLS was provided with \$45,000 to develop a long-term POC bottom of the watershed monitoring program which included selecting the 4 initial sites for monitoring as well as providing input and oversight on the RWSM and other RMP products. This planning and oversight included 6 in person work group meetings and monthly phone meetings. The monitoring program was successfully implemented in WY2012. No funds were requested for STLS management in 2012.

We are requesting \$20,000 to support STLS activities in 2013. Support will include quarterly STLS meetings to collaborate and coordinate WY2013 POC monitoring, provide updates and solicit input on RWSM, and EMC development. Funds will also include monthly phone conferences for brief updates and information sharing.

APPLICABLE RMP MANAGEMENT QUESTIONS

Level I RMP, Q3:	What are the sources, pathways, loadings, and processes leading to
	contaminant-related impacts in the Estuary?
Level II RMP, Q3C:	What is the effect of management actions on loads from the most
	important sources, pathways, and processes?
Level III SPL Q2:	What is the watershed-specific and regional total water flow, load of
	sediment, and load contaminants entering the Bay from the
	urbanized small tributaries and non-urban areas draining to the Bay
	from the nine-county Bay Area and are there trends through time?

Item #7 RMP Study #11	McKee and Hunt
STLS: Management Support for Spreadsheet Model Outreach	Page 2 of 2
and "Land Use" Based Monitoring	
Level IV STLS Q1: Which are the "high-leverage" small tributarie	

Level IV SILS QI.	which are the high-reverage small urbutaries that contribute of
	potentially contribute most to Bay impairment by pollutants of
	concern?

Level IV STLS Q2:	What are the annual loads or concentrations of pollutants of concern
	from small tributaries to the Bay?

BUDGET

Proposed Cost			
Task 1	Local STLS meetings	\$15,000	
Task 2	Phone conferences for product updates and review	\$5,000	
<u>Total</u>		<u>\$20,000</u>	

Nutrient Studies: Moored Sensor Monitoring Program Development, Algal Biotoxin Monitoring, Stormwater Nutrient Measurements, and Load Quantification

PROPOSED BY:David Senn (SFEI), James Cloern (USGS), Tara Schraga (USGS), Raphael
Kudela (UCSC), Martha Sutula (SCCWRP), Naomi Feger (SFRWQCB)

This proposal was developed in consultation with the Nutrient Stakeholder Advisory Group (SAG). The SAG met June 22, 2012 to review this proposal.

ESTIMATED COST: \$355,000 (\$320,000 after applying \$35,000 of unused 2012 USGS funds) **OVERSIGHT GROUP:** Nutrient Workgroup through the Nutrient SAG

TABLE 1: PROPOSED DELIVERABLES, BUDGET, AND TIMELINE

	Buc	lget	Des Data	
Deliverable	RMP	Other	Due Date	
Task 1 Project management	20K		Jan-Dec 2013	
Task 2. Moored sensor monitoring program development	200K	70K		
Task 2.1 Moored sensor platform selection and purchase			Jan-Feb 2013	
<i>Task 2.2</i> Sensor system calibration and basic dockside operation			Apr-May 2013	
<i>Task 2.3</i> Sensor deployment, operation, and maintenance at Dumbarton Bridge			Jun-Dec 2013	
<i>Task 2.4</i> Data analysis, QA/QC			Jun-Dec 2013	
<i>Task 2.5</i> Develop calibration, operation, and maintenance manual			Nov 2013	
<i>Task 2.6</i> Technical memo: data interpretation and recommendations for next steps with moored sensors			Mar 2014	
Task 3 Developing Solid Phase Adsorption Toxin Tracking (SPATT) as a Monitoring Tool for Microcystins and Related Toxins65Kin San Francisco Bay65K		-		
<i>Task 3.1</i> Monthly monitoring of algal biotoxins during Bay-wide monthly cruises and at fixed sites			Jan-Dec 2013	
<i>Task 3.2</i> Controlled experiments to calibrate biotoxin field sampling device			Jan-Aug 2013	
Task 3.3 Technical memo			Mar 2014	
Task 4 Stormwater nutrient monitoring in 6 Bay area catchments	40K	>300K		
Task 4.1 Field sampling and sample analysis			Nov2012- Apr2013	
Task 4.2 Data analysis and preparation of technical memo			June-Aug 2013	
Task 5 Technical report quantifying nutrient loads to the Bay and identifying data gaps		-	June 2013	
Nutrient/phytoplankton biogeochemical modeling (see CFWG modeling proposal				
Total	355K	>370K		

Background and Justification

San Francisco Bay has long been recognized as a nutrient-enriched estuary, but one that has historically proven resilient to the harmful effects of nutrient enrichment, such as excessive phytoplankton blooms and hypoxia. The published literature suggests that the accumulation of phytoplankton biomass in the Bay is strongly limited by tidal mixing, grazing pressure by invasive clams, light limitation from high turbidity, and potentially, in the North Bay, ammonium inhibition of diatom uptake of nitrate. However, evidence is building that, since the late 1990s, the historic resilience of the Bay to the harmful effects of nutrient enrichment is weakening (Cloern et al., 2007; Dugdale et al, 2007).

In response to the apparent changes in the Bay's resilience to nutrient loading, SFEI has been working with the San Francisco Bay Regional Water Quality Control Board and Bay area stakeholders to develop the San Francisco Bay Nutrient Strategy (http://www.waterboards.ca.gov/sanfranciscobay/water_issues/programs/planningtm dls/amendments/estuarynne.shtml). The goal of the Nutrient Strategy is to lay out a well-reasoned and cost-effective program to generate the scientific understanding needed to fully support major management decisions. The Nutrient Strategy has 6 main goals:

- 1. Define the problem: develop conceptual models for Bay segments that characterize important processes linking nutrient, biological responses, and indicators of adverse effects of nutrient over-enrichment
- 2. Establish guidelines (water quality objectives; i.e., assessment framework) for nutrients, including ammonium, focusing on the endpoints of eutrophication and other adverse effects of nutrient overenrichment;
- 3. Implement a monitoring program that supports regular assessments of the Bay;
- 4. Develop and utilize nutrient-load response models to support nutrient management decisions;
- 5. Evaluate control strategies to reduce nutrient inputs from wastewater treatment plants and other sources; and
- 6. Consider alternative regulatory scenarios for how to move forward with nutrient management in SF Bay.

This proposal to the RMP is requesting funds to support technical studies that directly address several goals of the Nutrient Strategy and key management questions, as noted within the description of each task. The Steering Committee provisionally allocated \$200,000 in 2013 for nutrient-related projects. In addition, the USGS had a one-time surplus in their funding, and is requesting \$35,000 less from the RMP in 2012, with the request that if possible those funds be allocated toward monitoring program development projects (e.g., Task 2 or Task 3).

While not discussed in this proposal, the CFWG modeling proposal has direct links to and benefits for nutrient work in the Bay. That proposal, with a requested budget of \$200,000 from the RMP and \$300,000 from other sources, aims to build the foundation

of a Bay-wide modeling program that can support modeling of bioaccumulative contaminants and nutrients/phytoplankton.

RMP Multi-Year Plan Priority Questions addressed by study proposal

- 1. Is there a problem or are there signs of a problem with respect to nutrient enrichment?
- 2. What are appropriate guidelines for assessing SF Bay health?
- 3. What is the relative contribution of nutrient loading pathways and how do loads vary seasonally and between Bay segments?

Goals of Nutrient Strategy addressed by study proposal

- 1. Document our current understanding of nutrient dynamics in the Bay, highlighting what is known and the crucial questions that need to be answered
- 2. Develop a monitoring program to support regular assessments in the Bay
- 3. Quantify nutrient loads to and important processes in the Bay

Task 2 Moored Sensor Monitoring Program Development

The indications of decreased Bay resilience to high nutrient loads noted above have come to the fore at a time when the availability of resources to continue assessing the Bay's condition is uncertain. Since 1969, a USGS research program has supported water-quality sampling in the San Francisco Bay. This USGS program collects monthly samples between the South Bay and the lower Sacramento River and measures salinity, temperature, turbidity, suspended sediments, nutrients, dissolved oxygen and chlorophyll a. The USGS data, along with sampling conducted by the Interagency Ecological Program, provide coverage for the entire San Francisco Bay –Delta system. The San Francisco Bay Regional Monitoring Program (RMP) has no independent nutrient-related monitoring program, but instead contributes approximately 20% of the USGS data collection cost. Thus, there is currently an urgent need to lay the groundwork for a locally-supported, long-term monitoring program to provide information that is most needed to support management decisions in the Bay.

While most of the historic and current data being generated by the USGS and IEP research programs are derived from ship-based measurements, there is a growing recognition that moored multi-sensor platforms can provide valuable temporallyintensive data. A number of large estuaries in the US (e.g., Chesapeake Bay, <u>http://mddnr.chesapeakebay.net/eyesonthebay/index.cfm</u>; Columbia River, <u>http://www.stccmop.org/datamart/observation_network</u>; Caloosahatchee Estuary, <u>http://recon.sccf.org/</u>) have well-established moored sensor networks that are integral components of monitoring efforts and provide a strong complement to ship-based monitoring. At a recent RMP-sponsored nutrient conceptual model technical meeting, the technical team recommended that pursuing a pilot project with moored sensors would be a valuable step that should be taken in the early stages of planning the next-generation monitoring program for the Bay. This group further advised that the RMP invest sufficiently in person power, beyond the cost of the hardware, to ensure the success of this effort and to allow ample time for sensor platform selection, operation

and maintenance, and data analysis, so that the effort contributes to monitoring program development.

Task 2 has been broken down into six subtasks (Table 1). Task 2.1 focuses on sensor platform selection. David Senn (SFEI) has already researched a variety of sensor platforms, and the LOBO system (http://www.satlantic.com/lobo) is our initial recommendation, because of its robust sensors (good track record based on other estuaries), including the considerable attention paid to minimizing biofouling. However other sensor platforms will be considered. The proposed LOBO sensor package includes conductivity, temperature, depth, dissolved oxygen, chl-a, turbidity, and nitrate, and both logs data and telemeters data via the cellular network to a manufacturer-provided web-interface for real-time data visualization (this will likely be most useful for tracking sensor drift or failure; however it could also be useful for detecting short-lived events, such as blooms, and triggering a focused field sampling campaign). The SUNA nitrate sensor (http://www.satlantic.com/suna) that is part of this package is a state of the art sensor and has sufficiently low detection limits (1-5 uM) relative to typical Bay conditions (~20-80 uM).

In Task 2.2, the LOBO system will be calibrated and tested in the lab, and then field tested for ~1 month at the Redwood City dock near USGS Menlo Park. In Task 2.3 the system will be deployed on a bridge piling at Dumbarton Bridge in June 2013, in collaboration with David Schoellhamer (USGS) whose group currently deploys and maintains turbidity, conductivity, temperature, and dissolved oxygen sensors at this site. The overlap in sensor capabilities is helpful because it will allow for continuous and colocated validation during the early stages of this pilot project. We will coordinate our maintenance schedule with Schoelhamer's group; they have agreed to collaborate on maintenance, which will allow both groups to leverage funds from various sources to support field work. Data will be collected continuously from June-December 2013, with on-going QA/QC (Task 2.4). Discreet water samples will be collected periodically (biweekly) adjacent to the sensor and measured for the suite of parameters to validate sensor operation. An operation and maintenance manual will be developed (Task 2.5). Finally, a technical memo will be produced that presents initial data analysis and synthesis, and just as importantly describes lessons learned during year 1 and recommendations for next steps with moored sensor applications (Task 2.6).

While this proposal requests funds for one year, it should be recognized that the moored sensor project is in reality a multi-year effort. In order to take full advantage of the investment, the costs to the RMP or to the eventual nutrient monitoring program will be on the order of \$115k/yr to continue this work in 2014 and beyond, and greater if more moored sensors are installed at other locations. (Note: Most of the projected cost for 2014 is salary for staff to maintain sensors, data QA/QC, and data interpretation. If the program eventually grows to more stations, SFEI staff effort will increase. However, we do not expect that, for example, a tripling in stations would translate into a tripling of staff cost; instead we believe that one person could eventually manage several moored sensor platforms located throughout the Bay. We contacted researchers in one

estuary that has 5 LOBO systems (<u>http://recon.sccf.org/</u>), and they reported that one person maintains and carries out data management for the entire network).

BUDGET – Task 2

USGS-Menlo Park will provide in-kind support equivalent to 20% of one FTE focused on sensor maintenance and calibration. USGS-Menlo Park will also contribute other in-kind salary support for project guidance, and in-kind support for analysis of discrete water samples for relevant parameters to validate sensor performance. USGS-Sac (Schoelhammer) will be providing in-kind support related to shared costs for maintenance/field work. We are also exploring options for funding support outside of the RMP through Nutrient Strategy collaborators, specifically in terms of purchasing hardware. However, those sources are uncertain and therefore the proposed budget is for the full cost of the project for 2013.

	RMP	Nutrient Strategy	USGS- Menlo Park	USGS-Sac	Total
Hardware and shipping	80K				80K
Personnel					
SFEI 50% FTE	89K				89K
5% Senn	16K				16K
5% Cloern (USGS)			14K		14K
5% Schraga (USGS)			10K		10K
USGS technicians			21K (20% FTE)	10K (10% FTE)	31K
Field/logistics	5K			10K	15K
Machining/engineering of deployment/retrieval of system	10K				10K
Sample analysis			5K		5K
Totals	200K	0	50K	20K	270K

Task 3 Developing Solid Phase Adsorption Toxin Tracking (SPATT) as a Monitoring Tool for Microcystins and Related Toxins in San Francisco Bay

Task 3 is also related to monitoring program development, focused on the detection of algal toxins produced by harmful algal blooms (HABs). There was broad agreement within the conceptual model technical team that increased frequency and magnitude of algal toxin monitoring measurements are one likely outcome of elevated nutrient loads to the Bay and Delta. The group further concurred that the development of sensitive tools for measuring phytotoxins should be a high priority for the Bay monitoring program.

Cyanobacterial blooms and their associated toxins have become increasingly problematic globally (Chen et al. 1993, Domingos et al. 1999, Lehman et al. 2005, Guo 2007, Paerl & Huisman 2008). Microcystis aeruginosa in particular is considered a cyanobacterial harmful algal bloom (CyanoHAB) organism because it can impede recreational use of waterbodies, reduce aesthetics, lower dissolved oxygen concentration, and cause taste and odor problems in drinking water, as well as produce microcystins, powerful hepatotoxins associated with liver cancer and tumors in humans and wildlife (Carmichael 2001). Extensive *Microcystis* blooms with toxin production occur during summer and fall in impaired waterways in Washington, Oregon and California (Gilroy et al. 2000, Johnston & Jacoby 2003) and Microcystis contamination has been documented at the marine outflows of the Klamath and San Francisco estuaries (Lehman et al. 2005, Fetcho 2007) as well as from river inputs to Monterey Bay (Miller et al. 2010). More recently, a SCCWRP study detected microcystins and/or anatoxin-a in 39 of 40 freshwater lakes and intermittently closed coastal lagoons tested within the coastal watersheds of five Southern California counties (Magrann, 2011). The recently documented direct impact to the threatened California Sea Otter (Enhydra lutris) has also promoted these blooms and toxins from predominantly a freshwater issue to potentially a land-sea problem, with concomitant risk because of the lack of monitoring in brackish and marine waters (Miller et al. 2010).

Until recently, *Microcystis* (and associated Cyanobacterial Harmful Algal Bloom (CyanoHAB) genera) blooms and microcystin intoxication were considered a public health issue solely of freshwater ponds, lakes, reservoirs, public water supplies and rivers; this assumption is reflected in the vast body of scientific literature available on potential public health risks from microcystin exposure in freshwater habitat. By comparison, monitoring of the freshwater-marine interface for similar ecological or public health risks has remained a low priority until very recently, despite observation of outflows of *Microcystis* and microcystin-contaminated fresh water to the ocean (Lehman *et al.* 2005; Tonk *et al.* 2007). Given the severe and ubiquitous nature of this problem in freshwater habitats and potentially coastal marine systems, surveillance and monitoring is critical. Traditional monitoring programs for phycotoxins typically rely on discrete sampling ("grab" samples) from a particular site or sites, sometimes augmented with automated sampling systems. Such methods are inherently biased if the sampling does not capture the spatial and temporal variability of the system due to behavioral adaptations of the algae such as vertical migration, hydrologic or circulation effects, and

ephemeral or episodic events. Furthermore, grab sampling may underestimate the presence of low levels of toxins if the sampling protocol does not include preconcentration and/or if the toxin concentrations are below the analytical limit of detection.

In response to this challenge, Dr. Kudela and colleagues at UCSC have been investigating the use of a passive sampling method, Solid Phase Adsorption Toxin Tracking (SPATT), to monitor microcystin (and other toxin) levels in seawater. SPATT was first proposed for HAB monitoring by MacKenzie et al. (2004), who developed this passive sampling device by placing SPATT resin, which binds an array of lipophilic algal toxins, within a polyester mesh bag. Over the last several years UCSC researchers have been further developing and applying SPATT for HAB detection in both marine and freshwater environments. Their results indicate that the sensitivity of this system is extremely high, which greatly facilitates source-tracking efforts. The researchers routinely detect biotoxins using SPATT when simultaneous point-sampling of water fails to detect the same toxins in a given waterway (Lane et al., 2010; Kudela, 2012).

Kudela and colleagues have conducted limited SPATT and grab-sampling within the Bay Delta and surrounding environment. Those data demonstrate that microcystins are present at moderate to high concentrations in source waters of the Bay (particularly the Delta, but also the ponds in the South Bay region; Figure 1). They have also tested SPATT in "flow-through" mode aboard the *R/V Polaris* during USGS cruises (Figure 2). Of particular concern, they have identified microcystins throughout the Bay during autumn, suggesting that toxins (but not necessarily cells) are being physically transported throughout the ecosystem.

Proposed Work Plan

Task 3 is divided into three subtasks. In Task 3.1, it is proposed to continue deployment of SPATT during USGS monthly cruises. As in past cruises (Figure 2), one SPATT will be deployed per basin in the surface-sampling flow-through system on the Polaris, totaling 5 SPATTs per cruise. In other watersheds, UCSC has successfully deployed SPATT from fixed platforms such as moorings (this has been done in the Delta, Alviso Slough and Pond A6, and throughout the Monterey Bay region). SPATT can easily be deployed up to 30 days, and require minimal handling for field personnel. SPATT can be stored indefinitely in the freezer (-80°C) and are routinely shipped through common carriers (including US Postal Service). In Task 3.1, SPATT will be deployed at both the Dumbarton Bridge and Benicia Bridge for periods of ~1 month, taking advantage of existing fixed monitoring programs. A similar effort in Pinto Lake, CA for a year was sufficient to develop statistical models relating toxin concentrations to environmental conditions (Kudela, 2012).

SPATT has now been extensively tested and applied for microcystin detection by UCSC, and there is widespread interest from the research and management community in deploying SPATT as part of existing monitoring programs. As part of the move toward more routine use of SPATT in monitoring programs, several issues related to SPATT

deployment and interpretation need to be addressed, such as how SPATT compare to ambient concentrations (calibration relative to in situ conditions), best practices for deployment/recovery and analysis of SPATT (i.e. length of deployment, analytical methods for toxin detection), and how SPATT compare with more mature passive samplers to quantify the partitioning coefficients and kinetics, and effects of flow and surface area. Some of these preliminary lab measurements have been conducted already (Kudela 2012) but UCSC has not extensively tested longer deployments or the partitioning in flow-through versus controlled volume situations.

In Task 3.2, controlled experiments will be conducted in the laboratory to better characterize partitioning of phytotoxins out of solution and into the SPATT during exposure in ship-board flow-through systems. Specifically, experiments will be carried out in simulated flow-through systems in which SPATT will be exposed to brackish water and seawater containing varying concentrations of a microcystin-RR. Microcystin-RR uptake will be quantified as a function of both dissolved concentration and exposure time. This "calibration" information will allow for more accurate back-calculations of average ambient concentrations in natural systems. In addition, a time-series of "bottle" experiments will be conducted during which SPATT will be exposed in containers holding seawater with known concentrations of microcystin-RR. SPATT will be removed at several time points and microcystin-RR uptake will be measured. This information will aid in characterizing the uptake kinetics of microcystin under conditions simulating longer term deployments at a single site.

In Task 3.3 a technical memo will be prepared that interprets the results from 2013 field sampling and the controlled experiments. The results of the field and laboratory studies are expected to inform future monitoring approaches in the Bay, and ultimately provide information to support management decisions related to HABs and biotoxins. It is anticipated that results will also be published as a journal article, to be submitted in the first half of 2014.

	RMP
Personnel	46.5K
Analyses, shipping, local travel, materials, misc.	18.5K
Total	65K

BUDGET – Task 3

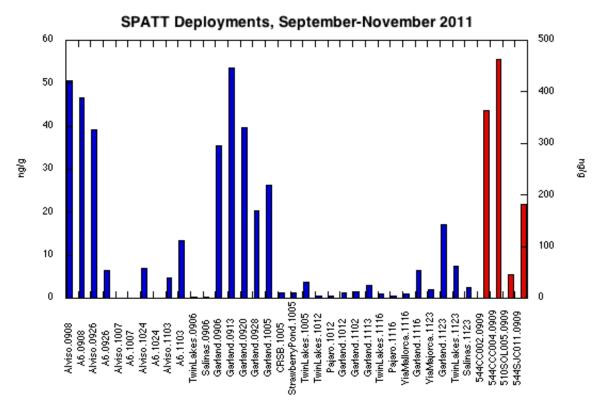
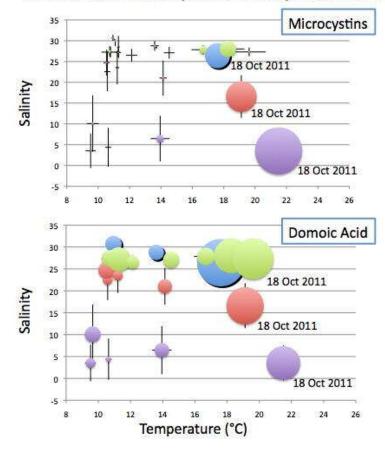


Figure 1. SPATT results from the South Bay region (left) and Monterey Bay (middle) in blue, and from the Delta (right; red) for microcystin-LR. While the Delta has orders of magnitude more toxin for equivalent sampling intervals, there are easily detected toxins throughout the other watersheds. The Monterey Bay toxin loads have been associated with California otter deaths, highlighting the negative impacts of these toxin levels.



SPATT concentrations plotted in Temperature-Salinity space



Figure 2. SPATT results from USGS deployments. Note that we detect multiple toxins from the same SPATT sampler, and that during some events there is extensive transport of both marine (domoic acid) and freshwater (microcystins) toxins throughout San Francisco Bay. Circle size indicates toxin concentration, bars represent standard deviation of T,S for each section.

Task 4 Nutrient stormwater sampling in 6 Bay area watersheds

Among the key objectives outlined in the March 2012 Draft Nutrient Strategy is the quantification of nutrient loads from main potential sources. Currently limited data exists to support the quantification of nutrient loads from storm water runoff. Developing accurate estimates for regional watershed loads to the Bay requires both acquiring empirical data from representative watersheds (for calibration/validation) and developing models to quantify loads across the region.

The Small Tributary Loading Strategy (STLS) led to monitoring of watersheds to quantify concentrations and loads of priority pollutants to the Bay during wet weather events. Study watersheds have been selected to represent the range of land use and land cover characteristics of the diverse watersheds draining to the Bay. The STLS is a multi-year effort, which studied 4 watersheds for up to 4 storms in 2011-2012 (3 watersheds monitored by SFEI, one by consultant). Six watersheds are proposed for monitoring in 2012-2013. Empirical data on flow and concentration will be collected and used to compute loads, and to calibrate spreadsheet models to estimate loads across the region. Development of the spreadsheet model is underway (Lent and McKee, 2011), but this model does not vet have capacity to predict nutrient loads. Although nutrients are not the main focus of the STLS, three nutrient analytes (nitrate, total phosphorous, dissolved orthophosphate) are among the current list of analytes because they are required as part of the Municipal Regional Stormwater Permit. However, other important nutrient analytes that are needed to provide a fuller picture of nutrient loads to the Bay are not funded because the permit does not require them (NH4, total Kjeldahl nitrogen (TKN)).

At the October 2011 RMP *Sources, Pathways, and Loadings Workgroup* meeting there was general agreement that the current suite of analytes should be augmented to include a full set of nutrient analytes, funds permitting. Adding these nutrient analytes, when teams are already mobilizing for the other contaminant sampling, is a wise investment, leveraging current funds being invested in this effort. NH4 and TKN were added to the list of analytes for the 2011-2012 sites, and that data has just become available and will be analyzed during Summer 2012 to inform potential sampling in 2013

We propose to collect samples for additional nutrient parameters at the six watersheds being sampled during the 2012-2013 rainy season. The proposed additional analytes are again NH4 and TKN. The combined suite of nutrient analytes matches the type of information being collected in the USGS monthly Bay surveys, and data being collected by POTWs over the next two years. Funds are also being requested for data analysis and preparation of a technical memo on all the nutrient data (including those already being measured by STLS), which will describe initial findings and make recommendations for field work in 2013-2014 and beyond. To the extent possible given budget constraints, a compilation of existing data on land use-specific runoff concentrations of nutrient forms will be compiled for a range of land uses through a collaboration with SCCWRP. The results from 2011-2012 and 2012-2013 wet weather seasons will provide important information for quantifying nutrient loads to the Bay

from urban runoff. It is roughly estimated that this proposed work is leveraging more than \$300,000 of field work and logistical support.

BUDGET – Task 4

	# of sites	# of sample s per site per storm	# of storm s	Total # of samples including QA/QC	Cost per analysis	Total per analyte
NH4+ and TKN analysis	6	4	4	110	\$113*	\$12,430
High turbidity filters –110 @\$13 each + shipping						\$1,950
Additional sampling effort						\$6,000
Contracting, project management						\$2,500
Data Management, QA/QC						\$2,500
Data analysis and preparation of technical memo						\$15,000
					Total	\$39,880

*TKN = \$50.40/sample; NH₄+ = \$62.40/sample

Task 5 Quantifying External Nutrient Loads and Data Gaps Analysis

Quantifying external nutrient loads to San Francisco Bay was identified as a highpriority funding item in the draft Bay Nutrient Strategy. Given that nitrogen (and to a lesser extent phosphorous) can experience multiple potential fates once entering an estuary, accurate load estimates are a pre-requisite for eventually developing reliable mass budgets and quantifying internal-Bay processes. In 2012, we proposed to develop spatially- and temporally-explicit estimates of nutrient loads to the Bay, and identify critical data gaps that contribute most to current uncertainty in total loads, speciation of those loads, and the relative importance of various sources. We proposed that this work would be distributed 40% and 60% between 2012 (\$20K) and 2013 (\$30K), respectively. Task 5 in this 2013 request is for the continuation and completion of that work.

A summary of external loads to the South Bay has already estimated by SFEI through funding from BACWA (McKee and Gluchowski, 2011). Task 5 (2012 and 2013) expands that loading work into the Central, San Pablo, and Suisun Bays, developing monthly, seasonal and annual load estimates, and exploring the importance of uncertainties in loading and nutrient speciation. The nutrient sources considered will include: POTW discharges; stormwater discharges; flows from the San Joaquin and Sacramento Rivers entering through the Delta; exchange across the Golden Gate; and direct atmospheric deposition. Unlike the South Bay, where loads from POTWs appear to dominate input of nutrients, other sources (e.g., flux through the Golden Gate; discharge through the Delta) likely contribute substantial proportions of the overall loads in the Central and North Bay. Loads from the Delta to the North Bay may be reasonably well-constrained, due to intensive monitoring in the region. Some of the funding in 2013 can be applied toward incorporating the historic and new discharge effluent data required under the Regional Water Board's March 2012 13267 Order to wastewater dischargers; that data will begin becoming available in the second half of 2012.

As part of identifying major uncertainties and data gaps, Task 5 will identify highpriority monitoring activities and special studies designed to better constrain nutrient load estimates.

	RMP
On-going data analysis, report preparation	30K

BUDGET – Task 5