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A Framework for Research Addressing the Role of Ammonia/Ammonium in the Sacramento-San Joaquin Delta and the San Francisco Bay Estuary Ecosystem

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INTRODUCTION

The Sacramento-San Joaquin Delta and the San Francisco Bay Estuary ecosystem has been drastically altered by human activity since at least the mid-1800s, leading to numerous physical, chemical and biological changes [e.g., see Healey (2008) and Healey et al. (2008) for overviews]. Recent concerns include (1) the precipitous decline of four fish species that inhabit the Delta (collectively referred to as the pelagic organism decline, or POD) and (2) apparent major shifts in community composition of algae and higher trophic levels in the Delta and downstream in Suisun Bay. The four POD species of concern are delta smelt (*Hypomesus transpacificus)*, longfin smelt (*Spirinchus thaleichthys*), threadfin shad (*Dorosoma pretense*) and striped bass (*Morone saxatilis*). One of the potential causes of the POD and the community changes is the discharge of increasing concentrations of a combination of ammonia (NH_3) and ammonium (NH_4^+) into the Delta ecosystem and eventually into Suisun Bay. For brevity, we hereafter abbreviate the ammonia/ammonium combination as ammonia/um.

To address the potential for adverse effects of ammonia/um on the Bay-Delta ecosystem, a ammonia/um workshop was convened by the CALFED Science Program on 10 and 11 March 2009 in Sacramento, California. We participated as an expert panel and were charged with preparing a framework for research that will address the role of ammonia/um in the Bay-Delta ecosystem.

In this report, we present our perception of the major concerns related to ammonia/um in the Bay-Delta ecosystem; a conceptual framework of the major physical, chemical and biological drivers controlling the ecosystem (including POD populations); our assessment of the major research needs; and concluding remarks. For some needs, we recommend specific types of research and provide literature citations; however, for other needs we recommend general approaches for which targeted research will be determined later. In many ways, Bay-Delta research is breaking new ground and will establish a template for future research and management decisions, rather than relying on paths trodden by others.

MAJOR CONCERNS RELATED TO AMMONIA/UM

Based on presentations and discussions at the workshop, we perceive two major concerns related to ammonia/um in the Bay-Delta ecosystem. First, the precipitous POD might be caused directly by toxicity of ammonia/um to the four fish species. Second, the Bay-Delta ecosystem is rapidly progressing through major food web shifts that include undesirable changes in community structure and productivity of the primary producers (phytoplankton and aquatic macrophytes) and intermediate consumers (e.g. zooplankton, larval fish). Those changes might be caused by ammonia/um and might be altering nutrient cycling and the physical-chemical habitat in the Delta, with concomitant but different ramifications downstream in Suisun Bay. Alternatively, ammonia/um enrichment might have no detrimental effects, and other drivers are responsible for the biogeochemical and trophic changes. Underlying these major concerns are several human activities, including (1) upstream discharges of ammonia/um-containing municipal wastewater [e.g., from Sacramento Wastewater Treatment Plant (SacWTP) into the Sacramento River], and (2) large-scale pumping of water from the Delta.

Conceptually, these two major concerns related to ammonia/um (the POD and the food web changes) are different yet possibly interrelated issues. For example, alternate hypotheses include: (1) current ammonia/um concentrations might not cause toxicity to any of the POD organisms, but ammonia/um-induced changes in phytoplankton community structure and productivity might be a major cause of the POD through cascading trophic interactions; (2) conversely, ammonia/um might not directly alter the structure and productivity of the phytoplankton community, but ammonia/um might be toxic to food organisms (e.g., invertebrates) on which POD organisms depend during at least part of their lives; or (3) ammonium enrichment is not a prime factor responsible for the trophic and biogeochemical changes.

In addition to ammonia/um, numerous other anthropogenic toxins (e.g., pesticides, metals) known to contaminate the Bay-Delta estuary and "natural" toxins (e.g., cyanobacterial exudates released by expanding *Microcystis* blooms) might directly affect POD organisms and/or decrease phytoplankton and/or zooplankton productivity. Furthermore, invasions of alien herbivores [e.g., overbite clam

(*Corbula amurensis*) and Asian clam (*Corbicula fluminea*)] during the past several decades and/or expansion of invasive aquatic macrophytes [e.g., Brazilian waterweed (*Egeria densa*)] might be the major cause of declining standing stocks of phytoplankton. Finally, export of Delta water altered hydrologic conditions, and temperature increases accompanying recent climate changes might be major physical factors controlling the Bay-Delta estuarine communities (including populations of the POD organisms).

Based on presentations at the workshop and the background material provided to us, it does not currently appear possible to conclusively reject any of the proposed explanations for the POD and the food web changes. In fact, the multiplicity of diverse potential drivers in this ecosystem needs to be placed into a conceptual framework that could help guide research in the Bay-Delta estuary ecosystem and eventually lead to well-grounded management decisions about ammonia/um.

CONCEPTUAL FRAMEWORK

Potential drivers of water quality and the structure and function of the Bay-Delta ecosystem include climate, hydrology (including water withdrawals and flow modifications), human activity, loadings and types of nutrients (mainly N and P, from anthropogenic and natural sources), loadings and types of contaminants (including NH_3/NH_4^+, NO_2^-, metals, pesticides and "natural" toxins), sediment loadings, light, and food web processes (including trophic interactions, with special emphasis on invasive species). These factors are interrelated in a complex web of physical, chemical and biological processes shown conceptually in Figure 1. Climate and hydrologic variability are closely related factors that, in conjunction with human activity, influence and to varying degrees control many of the other drivers (e.g., delivery of nutrients and contaminants, changes in residence time). Therefore, climate/hydrologic variability and human activity are placed on the left in Figure 1, with consequences of those factors cascading from left to right through all the other drivers and ecosystem components. The endpoints of major concern in this framework are changes in the Bay-Delta food web and populations of the POD organisms, as shown on the right in Figure 1. For simplicity, arrows indicating some potential interactions (e.g., from human activity) are not included.

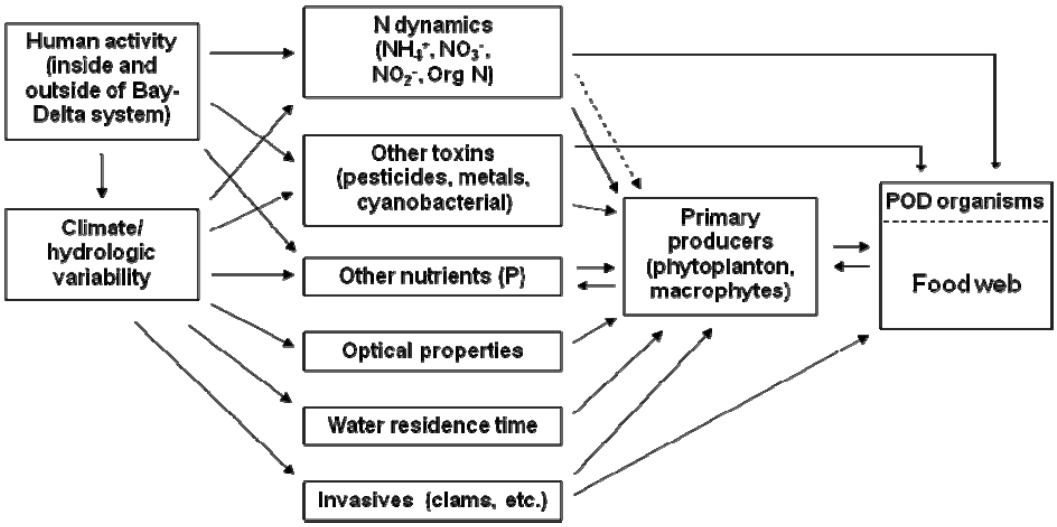

Figure 1. Conceptual framework of major drivers of water quality and ecosystem structure and function, and their relationships to the food web and POD organisms, in the Sacramento-San Joaquin Delta and Suisun Bay ecosystem. The solid arrow from the N dynamics box to the primary producers box indicates traditional processes associated with nutrient supply and uptake; the dashed arrow indicates the proposed inhibitory/competitive effect of NH_4^+ on uptake of NO_3^- by diatoms.

Complexity in the Bay-Delta Ecosystem

The Delta-Suisun Bay component of the San Francisco Bay ecosystem is a hydrologically, biogeochemically and trophically complex continuum. Hydrologically, this region is characterized by dynamic gradients attributable to interannual, seasonal, and episodic (storms, droughts) variability in freshwater discharge and tides. This variability controls water residence time, which influences flushing and nutrient and sediment delivery, which in turn control the magnitude and location of phytoplankton composition and standing crop. The ramifications of these interactive controls on the phytoplankton community have been discussed by Jassby (2008). Grazing by benthic and planktonic fauna exerts additional controls on the seasonal patterns and magnitudes of primary production and phytoplankton biomass. Lastly, light availability and the proportions of N and P loading play roles in controlling the magnitude and composition of the phytoplankton community. These drivers appear to interactively control phytoplankton production, biomass and composition (Jassby 2008).

Flow and residence time exert strong influences on the Delta and Suisun Bay. In the Delta, summer periods of relatively low flow and long residence time, in combination with adequate nutrient (N and P) supplies, favor the development and persistence of nuisance *Microcystis* blooms that produce toxins and alter food webs. These cyanobacterial blooms have increased in recent years, in part due to persistent drought conditions (poor flushing, long residence time), a recent warming trend, and increasing nutrient (especially N) loads. These factors synergistically favor cyanobacterial dominance (Paerl and Huisman 2008, 2009).

Because the dominant cyanobacterial genus in the Delta (*Microcystis*) does not fix N_2, these increasingly more common and extensive cyanobacterial blooms indicate sufficient and possibly excessive N loading to the Delta. Increases in NH_4^+ concentrations specifically might exacerbate this situation. Compared to NO_3^- and N_2 as N sources, NH_4^+ produces the highest growth and primary production rates for *Microcystis aeruginosa* and other cyanobacteria (*Aphanizomenom flos-aquae* and *Anabaena flos-aquae*) in laboratory studies (Ward and Wetzel 1980). The use of NH_4^+ might be particularly important for sustained cyanobacterial growth at low light intensities, because it is energetically favored compared to other dissolved inorganic N sources.

Microcystis is sensitive to salinity in excess of ~2 ppt (Paerl 1983; Sellner 1997). Therefore, *Microcystis* and possibly other nuisance cyanobacterial taxa (e.g., *Anabaena, Aphanizomenon*) are confined to the low-salinity waters of the Delta. However, in places where filter-feeding mussels and clams overlap with habitat suitable for *Microcystis* (i.e., low salinity), the presence of these invertebrates might enhance bloom formation by selectively rejecting large *Microcystis* colonies. That grazer selectivity can give *Microcystis* a grazer-resistant, competitive advantage over other phytoplankton, as Vanderploeg et al. (2001) reported for zebra mussels (*Dreissena polymorpha*) in the Great Lakes.

Downstream in Suisun Bay, phytoplankton composition, productivity, standing crop, and bloom dynamics appear to be closely controlled by freshwater discharge and residence time. The long-term data in Jassby (2008) indicate that in years when discharge is low, spring diatom-dominated phytoplankton productivity and blooms tend to be relatively small, whereas in high-flow years, these blooms are more prominent. Summertime phytoplankton productivity and bloom formation tend to be less coupled to flow conditions. High flow delivers higher nutrient (N and P) loads, which may increase the probability of phytoplankton blooms. Diatoms have fast growth rates and may be particularly good competitors during high flows with concomitant short residence times, when their fast growth rates can offset high flushing rates. In moderate flows, chlorophytes and cryptophytes become more competitive, whereas low flows with concomitant longer residence times allow the slower-growing cyanobacteria, non-nuisance picoplankton, and dinoflagellates to contribute larger percentages of the community biomass. These spatially and temporally-variable patterns of phytoplankton composition are typical of many estuaries [e.g., Chesapeake Bay, Maryland; Neuse-Pamlico Sound, North Carolina; Narragansett Bay, Rhode Island; Delaware Bay, Delaware] (Adolph et al. 2006, Valdes-Weaver et al. 2006, Paerl et al. 2009). This scenario might also apply to Suisun Bay and downstream San Francisco Bay (e.g., Lehman et al. 2008).

The relationship between flow, nutrient loading, and phytoplankton community composition and biomass in Suisun Bay is complicated by several other factors, including turbidity (transparency) and

grazing. Turbidity and resultant light limitation play an important role in controlling photosynthetic activity and hence primary production (Cloern 1999) and might also select for motile phytoplankton (flagellates, bouyant cyanobacteria) capable of remaining in the upper water column. Grazing, especially by invasive benthic infauna (e.g., clams) can exert strong "top down" controls on phytoplankton in northern San Francisco Bay, including Suisun Bay (Cole et al.1992, Jassby 2008). In addition, different inorganic N species might influence the composition and biomass of phytoplankton blooms. In particular, NH_4^+ "inhibition" of NO_3^- utilization when NH_4^+ concentration exceeds 4 μM has been proposed as a mechanism suppressing spring diatom blooms (Dugdale et al. 2007). Although such a mechanism might influence community composition, the dominant controls on productivity and biomass appear to be freshwater discharge (residence time) and grazing (Jassby 2008).

State Changes in the Bay-Delta Ecosystem

There is compelling evidence that recent anthropogenically imposed changes in the San Francisco Bay ecosystem have had dramatic impacts on production and composition of phytoplankton and macrophyte communities. The most notable changes are invasive benthic filter-feeding species and hydrologic modifications (i.e., water withdrawal and channelization). These changes appear to have led to biological and nutrient-cycling modifications and alterations that warrant being called "state changes". For example, the recent establishment and persistence of nuisance *Microcystis* blooms in the Delta seem to fit the "state change" paradigm, because dramatic changes in C and nutrient cycling as well as food web dynamics have accompanied the transition from a largely eukaryotic planktonic, grazer-oriented food web to one in which benthic/sedimentary processes are more dominant. Another example is the "top down" effects that invasive benthic filter feeders have had on phytoplankton communities in San Francisco Bay. In addition to having caused major changes in phytoplankton community biomass and composition, the presence of these benthic filter feeders has led to altered flowpaths, cycling and fates of C and nutrients. Such modifications appear to qualify as "state changes". However, more gradual changes might modulate these "state changes" or qualify as major effectors of state changes. These include (1) changes in nutrient input amounts and ratios, (2) changes in climate, including temperature and precipitation, and (3) within-system feedbacks due to altered nutrient and trophodynamic changes resulting from changes in the phytoplankton (i.e., cyanobacteria) and macrophyte "players" that have become more dominant in the system.

Changes in nutrient-loading amounts and ratios (nutrient stoichiometry) over time might typify the latter group of more gradual, chronic effectors. For example, increased loading of N (and specifically NH_4^+) that has impacted this region could have caused a shift in the Delta phytoplankton community toward *Microcystis* in a relatively short period (~5 years). This shift, in turn, has led to major modifications in C and nutrient cycling as well as trophodynamics. Another example is temperature shifts. Warming has been documented for this region, and warming has been linked to a greater preponderance of cyanobacterial bloom species (Paerl and Huisman 2008, 2009). Therefore, more gradual physical, chemical and biotic changes might play central, integral roles in "state changes" that have been observed in this region over the past several decades.

The Delta once included extensive tules, which are marshes composed primarily of emergent aquatic macrophytes (e.g., *Scirpus* and *Schoenoplectus* spp.). Less than 5% of this native marsh type currently remains, although portions still occur in the Delta and especially in the Suisun Marsh (Kimmerer et al. 2008). These marshes undoubtedly had a critical impact on multiple ecological features of the larger estuarine ecosystem, including high plant productivity, nutrient retention, nutrient and material export (e.g., dissolved organic matter), water-flow modification, and habitat for numerous wetland invertebrates, amphibians, reptiles, fish, and waterfowl.

The reconfiguration and partitioning of the Delta landscape over the last 150 years has drastically altered hydrologic flow patterns that connected the marsh communities and their influences with down-gradient bays. A different type of macrophyte community is now established and increasing in areal extent, including the invasive Brazilian waterweed and water hyacinth (*Eichhornia crassipes*) (Kimmerer et al. 2008). The water hyacinth is a free-floating plant (not rooted) that can accumulate massive amounts

of biomass at the water surface, has formidable nutrient retention capabilities (both N and P), and can create a low-light, at times hypoxic, environment in the water column beneath it (Mitsch 1977, Mitsch and Gosselink.2000). Brazilian waterweed is a submersed plant that can grow rooted in the sediments or unattached, and can also form dense surface mats. Both of these plant species differ substantially from the native, emergent macrophytes that once dominated Delta marshes. However, their effects on nutrient sequestration and transformation as well as biogeochemical processes associated with benthic sediments are largely unknown. Those biogeochemical processes could influence and facilitate trophic changes in down-gradient bays. Additionally, these new plant communities appear to foster growth of invasive, predatory fishes, such as black bass (Kimmerer et al. 2008). Therefore, the new plant communities might not only impact overall nutrient export, including inorganic and organic forms of N and P, but might also create favorable habitat for invasive fishes that might out-compete valued native species.

MAJOR RESEARCH NEEDS

We are impressed by the extensive amount of research that is being conducted on a wide variety of topics related to the Bay-Delta ecosystem. Many of the important research needs have been or currently are being addressed, and it is evident that considerable thought has been invested in the selection and design of those projects. Herein, we suggest several additional research areas and considerations that could help elucidate important processes and help managers make better-informed decisions about key human activities influencing the Bay-Delta ecosystem, including upstream discharge of ammonia/um into inflowing waters and large-scale pumping of water from the Delta. We present these suggestions in the framework of the three major topic areas discussed at the workshop: (1) sources, concentrations, fate and transport of nutrients, (2) food web effects, and (3) toxicity. Additionally, we have identified modeling as a fourth major topic area. However, before listing these additional research areas, we address an important concern that eclipses these individual topic areas.

We believe that the most important gap to be filled in the Bay-Delta research program is the development of an overarching, integrative model of the major drivers controlling the Bay-Delta ecosystem. This type of model would be instrumental in guiding current and future research, integrating the wide variety of information that is currently available and being generated and predicting outcomes of potential management actions and other changes upstream and in the Bay-Delta region. Development of this model will be time-consuming and challenging, but the outcome could be very useful.

We recommend developing as holistic a model as possible. However, we also recognize that important limitations related to availability of information, time, and money will pose major constraints; and at some point, the modeling effort will have to be ended conservatively – focusing only on the perceived most important processes, at workable spatial and temporal scales. Because of the complexity of the Bay-Delta ecosystem and its major drivers, we believe the model will have to be spatially and temporally explicit; and it will have to be flexible enough to be able to accommodate future stressors and changes. Major aspects of the model should include hydrology, the biogeochemistry of N and P, and trophic structure and function in the Bay-Delta ecosystem.

Jassby (2008) provides a rationale for this type of integrative model; and Figure 1 presents a preliminary conceptual framework. Based on existing models of hydrology, climate change, transport and transformations of nutrients and other aqueous contaminants, particle loading and transport in water, light penetration, and other physical-chemical processes, many of the interactions shown on the left half of Figure 1 could be quantitatively incorporated into the overarching model. However, some of the biological processes represented mostly on the right side of Figure 1 might have to be modeled only qualitatively, because of the complexity of the ecosystem structure and function, lack of sufficient detailed knowledge about the processes, and the need to retain manageable size, structure and cost of the model. Because of the underlying importance of hydrology to all the ecosystem processes in the Bay-Delta region, it might be advisable to use a currently available hydrologic model as a platform into which the other ecosystem processes and components can be incorporated.

Of prime importance to developing an overarching, integrative model is an integration of the

understanding of the roles of hydrology, nutrients, and herbivory in the temporal dynamics of phytoplankton production and community composition, including positive and negative feedbacks between the Delta and Upper Bay. Jassby (2008) provides a good start on this integration. Beyond that higher-level effort, crucial knowledge that needs to be generated and/or expanded to feed the model include: (1) an analysis of sources (exogenous and endogenous), sinks, and transformations of N along the Delta-to-Bay continuum, and controls on those pools and processes; (2) an understanding of factors that control POD populations, including various forms of N and a combination of other stressors, including chemicals, food availability and hydrology (including water-withdrawal systems); and (3) field observations of POD species and other potentially interacting and/or sensitive taxonomic groups. More specific recommendations about the types of research projects that could fill these research gaps are presented in the following sections, arranged by major topic area.

Modeling

Research Topic 1 – Modeling analysis of historical controls on phytoplankton populations:

Several co-occurring and correlated changes have occurred in the Delta and Suisun Bay. For example, changes in freshwater discharge have affected delivery of ammonia/um and other N species and P, and they have changed water residence time. The locations, magnitudes, and species composition of phytoplankton blooms in these systems appear to be related to these changes.

Which is (are) the main driver(s) of phytoplankton productivity, biomass, and species composition? Do these two drivers interact synergistically (i.e., in a more-than-additive interaction)?

Integrative (hydrology, biogeochemistry and food web dynamics) analysis and modeling are needed to address these questions. It is important to examine effects of these drivers on all three indices (phytoplankton productivity, standing crop, and species composition) because results might differ for these endpoints, especially if grazing exerts a major control on standing crop.

Modeling efforts should be directed toward distinguishing the influence of changes in nutrient loading (i.e., ammonium increases) and its effects from changes in freshwater discharge/flow (including residence time) and grazing, on cyanobacterial blooms in the Delta and on downstream blooms of phytoplankton (diatoms and other algal groups) in Suisun Bay. Additionally, the potential importance of those effects should be ranked for a variety of plausible combinations of hydrologic and nutrient-loading conditions.

Sources, Concentrations, Fate and Transport of Nutrients

Research Topic 2 – Sources and fates of N and P:

The central question for this research topic is: What is the role of the SacWTP versus other external and internal sources in controlling (a) ammonia/um concentrations and N dynamics in the Delta and Suisun Bay and (b) exports downstream? Although the emphasis is on N for concerns about the POD and food web changes in the Bay-Delta ecosystem, P loading should also be included in this analysis because future controls on N loading could alter nutrient stoichiometry, fluxes and limitations.

Discharge of N from SacWTP is well-documented, but there is a need to quantify other current and potential future sources of N, as well as the importance of internal N dynamics in controlling ammonia/um concentrations and the N species available to algae in the Delta, Bay and downstream ecosystems. These are needed to help determine the role of N (particularly ammonia/um and NO_2^- concentrations) in the POD and in observed changes in phytoplankton production and species composition [see Sommer et al. (2007), Jassby (2008), Lehman et al. (2008)].

Although SacWTP is a large source of N, particularly NH_4^+, other significant sources such as drainage from wetlands, urban stormwater runoff, and agriculture might exist. Furthermore, N transformations within the Delta, including mineralization of organic N, nitrification, assimilation of NH_4^+ and NO_3^-, denitrification, and N-fixation, might (a) mask the effect of specific sources and (b) control the concentrations and speciation of N. Therefore, a more specific question that should be addressed is: Is ammonia/um behaving largely conservatively in the Delta, or is it highly dynamic? If the

latter is correct, a better understanding is needed of sources (external and internal), N cycling rates, and associated lag times in transport of N along the major axes in the Delta (the Sacramento and San Joaquin Rivers), as controls on ammonia/um and exports of all forms of N to Suisun Bay and downstream.

Coupled hydrodynamic and biogeochemical models, such as the DSM-2 model, may be useful with current parameterization and calibration to define upper and lower bounds on effects of N loads and cycling. However, additional field measurements of N biogeochemistry and transport will be crucial to more accurately calibrate such models. In particular, the existing field data on N cycling rates within the various Delta habitats and over different seasons do not appear to be adequate.

Three approaches for future research to fill these data gaps seem promising. First, longitudinal transects of N concentrations (total ammonia, NO_3^-, dissolved and particulate organic N), important ancillary parameters (dissolved oxygen, pH, PO_4, and chlorophyll concentrations), stable isotope composition of ammonia/um (^{15}N), NO_3 (^{15}N and ^{18}O), dissolved and particulate organic matter (^{15}N, ^{13}C, ^{34}S), and N_2 concentrations [or N_2:Ar ratios, which are potentially useful for determining denitrification rates downstream from large point sources of N, as demonstrated by Laursen and Seitzinger (2002) and McCutchan et al. (2003)] should be established. These transects should be monitored during different seasons and under different hydrologic conditions, and would provide important diagnostic information about inputs of external sources of N as well as the importance of internal N cycling in controlling N concentrations within the Delta. The stable isotope measurements should be modeled after the recent work of Carol Kendall (U.S. Geological Survey) in the San Joaquin River system.

The second approach involves high-frequency measurement (e.g., hourly) of N species (particularly total ammonia and NO_3^-), together with the ancillary parameters listed above and perhaps N_2:Ar ratios, at several key locations within the Delta (e.g., immediately downstream from SacWTP, in the Sacramento River just upstream from its confluence with the San Joaquin River, and in Suisun Bay). These measurements should be made over extended periods (several weeks) during each season or under different hydrologic conditions, to provide important information on short-term temporal dynamics and the role of processes involving diurnal dynamics (phytoplankton uptake).

The third approach is direct measurements of such potentially important processes as uptake of NH_4^+ and NO_3^-, nitrification, mineralization of sediment and suspended organic matter that release NH_4^+ to water, denitrification, and N-fixation should be made in key Delta subsystems during different seasons, to provide the information needed to parameterize the large-scale biogeochemical models. These direct measurements can be made using standard techniques (concentration changes during incubation of water or sediments) as well as ^{15}N-tracer addition approaches.

Research Topic 3 – Nutrient dynamics in stands of aquatic macrophytes:

A more comprehensive approach is needed to investigate the roles of expanding populations of aquatic macrophytes in nutrient retention and export, as distinguished from the impact of N input from the SacWTP.

Quantification of changes in areal extent of the macrophyte beds can be accomplished through low altitude aerial photography coupled with ground-truthing of specific types of macrophyte taxa and GIS analysis. Color-Infrared (CIR) photographs acquired from relatively low-altitude flights will provide appropriate resolution and scale for plant community identification and tracking of areal changes over time [e.g., see methods in Cherry et al. (2009)]. Annual flights or twice per year flights should provide the frequency necessary to quantify changes in macrophyte community extent over multiple years.

Tracking nutrient changes upgradient, through, and downgradient from selected macrophyte beds in comparison with longitudinal transects through water without macrophytes will provide preliminary information on the effects of macrophyte stands on nutrient transport and transformation. Measurements of nutrients should include those described in Research Topic 2 above, with emphasis on dissolved organic and inorganic N forms and PO_4, but also including particulate and dissolved organic C. An appropriate complement would be an experimental or manipulation approach contrasting treatments with and without macrophytes, with controlled, unidirectional water flow.

Food Web Effects

Research Topic 4 – Links between nutrient processing and phytoplankton populations:

As an extension of Research Topic 2, rate processes of *in situ* N cycling, including nitrification, N-fixation, denitrification, and N regeneration need to be linked to other processes (as rates), including primary production by phytoplankton and rates of change of their biomass in the Delta and Suisun Bay. To that end, phytoplankton community composition in the Delta and Suisun Bay should be analyzed in addition to Chl *a*, as biomass responses to nutrient inputs and physical drivers. Because enumeration of individual species is time-consuming and tedious, fluoroprobe and group-specific measurements including diagnostic pigments will provide more cost-effective indices of phytoplankton community composition (Pinckney et al. 2001, Valdes-Weaver et al. 2006). Nutrient cycling measurements should accompany analyses of phytoplankton community structure, as part of recommended nutrient and phytoplankton monitoring programs. The outcome will help parameterize the modeling to be conducted under Research Topic 1.

Research Topic 5 – Bottom-up controls of higher trophic levels and POD organisms:

In the Delta, cyanobacterial blooms and expansion of aquatic macrophytes have occurred contemporaneously and contiguously. These changes probably have impacted food web dynamics, including a possible shift from a planktonic-based to a benthic-based food web.

Do the cyanobacterial blooms and macrophyte stands act as nutrient filters/transformers?

How have these changes affected higher trophic levels and POD organisms?

The rationale for these questions is addressed in part in above text on *Microcystis* use of ammonium and phosphate (see Complexity in the Bay-Delta Ecosystem section) and the role of Brazilian water weed and water hyacinth in nutrient uptake and as habitat for invasive fishes (see State Changes in the Bay-Delta Ecosystem section). Research approaches to answer these questions would overlap with the approaches discussed in those sections.

Research Topic 6 – Effects of climate change on phytoplankton:

Predicted climate-change scenarios include increases in temperature, intensification of spring freshwater discharge and flows, and decreased summer/fall flows.

How will these anticipated changes impact the expansion of cyanobacterial blooms in the Delta and possibly spring and other seasonal phytoplankton blooms in Suisun Bay?

This research question should be tested experimentally and by analysis of the historical record, and a sensitivity analysis should be conducted by adding a temperature component to the hydrology and nutrient-loading model developed for Research Topic 1.

Research Topic 7 – Thresholds for phytoplankton responses to nutrients:

Recent "state changes" in chlorophyll *a* (Chl *a*) concentrations and primary productivity in Suisun Bay appear to be more closely related to the recent rapid expansion of invasive filter feeders (i.e., clams) and relatively rapid changes in freshwater discharge, than to the more gradual increases in NH_4^+ concentrations/loading during the past few decades.

Could threshold responses to changes in nutrient (N and P) loading also play a role in these recent "state changes" -- instead of or in addition to the roles of invasive filter feeders and hydrologic changes?

Research Topic 8 – Lag times in phytoplankton bioassays:

Lag times in the growth response in phytoplankton bioassays of Bay-Delta waters are typically 1 to 2 days (personal communication, P. Lehman, California Department of Water Resources, Sacramento, CA). Dugdale et al. (2007) attributed this delayed response to NH_4^+ inhibition of (or competition with) NO_3^- utilization by diatoms, which continues until the NH_4^+ concentration is decreased to less than approximately 4 µM by phytoplankton uptake. However, this type of lagged response is also typically observed in other phytoplankton bioassays (c.f. Rudek et al. 1991, Fisher et al. 1992), regardless of

whether diatoms are a significant and/or the dominant phytoplankton group.

Is the delayed growth response in Bay-Delta phytoplankton bioassays caused by "container effects" and/or inherent lag times for phytoplankton response, or by NH_4^+ inhibition of (or competition with) NO_3^- utilization?

Research Topic 9 – Inhibition versus preferential uptake of ammonium:

The following questions are an extension of Research Topic 8.

How can NH_4^+ inhibition of NO_3^- uptake be distinguished from preference for NH_4^+ over NO_3^- uptake/utilization, especially at low to intermediate NH_4^+ concentrations?

From an ecological perspective (especially growth rates of different phytoplankton taxa), does it matter whether the mechanism is "inhibition" or "preference" for NH_4^+?

Per amount of N assimilated, is there a change in productivity, C:N uptake ratios, and growth per unit N uptake when phytoplankton are growing on NH_4^+ versus NO_3^-?

Are growth responses simply determined by the relative concentrations of the various N forms, rather than the absolute concentration of NH_4^+?

If this phenomenon only affects the spring diatom bloom, what are the trophic ramifications?

Nutrient addition (and dilution) bioassays, in which N and P are manipulated under natural irradiance and mixing conditions need to be conducted in water samples from the Delta and Suisun Bay (i.e., not only testing NH_4^+ inhibition of NO_3^- uptake). Response parameters should include Chl *a*, species counts, and/or the parallel use of quantitative diagnostic techniques, such as "phytoprobe" and indicator pigment techniques (Pinckney et al. 2001). These types of bioassays will allow investigators to examine phytoplankton biomass and compositional responses to changes in nutrient concentrations, and could be used to "parameterize" and/or validate models of those processes.

Research Topic 10 – Depression of phytoplankton biomass:

During the period when Dugdale et al. (2007) suggested NH_4^+ inhibited NO_3^- uptake by diatoms, were benthic clam filtration rates high enough to depress the phytoplankton (including diatom) biomass -- as suggested in Cloern (1982), Thompson and Nichols (1996) and Jassby (2008).

Phytoplankton production and consumption (grazing) rates need to be compared in order to evaluate the importance of these effects as controls on the spring phytoplankton bloom. The comparison in Wilkerson et al. (2006: p. 414) was not sufficient.

Toxicity

Research Topic 11 – Sensitivity of POD organisms to ammonia/um:

Relatively little is known about the toxicity of ammonia/um to the POD fishes. Aside from the recent acute toxicity tests conducted with delta smelt exposed to ammonia/um (Werner et al. 2009), only one other POD species that has been tested with ammonia/um – striped bass in an old study (Hazel et al. 1971) that should be repeated with larval fish.

First, a complete set of acute toxicity data should be generated (i.e., 96-h ammonia/um LC50s for all four POD fish species) for all four POD fish species, using standard methods (e.g., USEPA 2002) so the sensitivities of those fish can be directly compared to the sensitivities of other fish and aquatic invertebrates. Second, chronic toxicity data should be generated for at least some of the POD fish species, under standard "unstressed" conditions and under stressed conditions. If feasible, the species having the lowest 96-h ammonia/um LC50 should be tested first, followed by other species as availability of fish, time, and resources allow. A relatively easy and relevant stressor to use during the chronic toxicity tests would be continuous swimming in a water current in a circular tank at, for example, ~1 body length/sec. The chronic effect concentration for swimming fish could be compared to the chronic effect concentration for fish tested concurrently in calm water, to determine whether stressed fish are considerably more sensitive to ammonia/um than unstressed fish [as suggested by Randall and Tsui (2002)]. Because these would be long-term toxicity tests, the effect of total starvation on ammonia/um

toxicity could not be tested.

Although salinity, temperature, feeding level and/or other types of stressors besides swimming could be varied in a more extensive ammonia/um toxicity testing matrix, the expenditure of resources needed to complete such a large testing program would not be justified unless the results of initial chronic tests discussed in the previous paragraph demonstrate that at least the most sensitive POD fish would be affected at ambient ammonia/um concentrations in areas of the Bay-Delta ecosystem that it inhabits. Unless the exposure concentrations exceed the effects concentrations considerably, disagreement might ensue about the relevance and interpretation of any laboratory toxicity test results. Additionally, the presence of numerous other anthropogenic and "natural" toxins and other stressors will increase the difficulty of interpreting the ammonia/um toxicity results, especially because relatively little is known about joint toxicity. Therefore, overemphasis on laboratory studies at the expense of conducting fewer field monitoring studies (see Research Topic 13) might not be cost-effective.

Research Topic 12 – Cyanobacterial toxins:

Although Lehman et al. (2005) found no evidence to strongly support the hypothesis that cyanobacterial toxins are adversely affecting POD species or other components of the Bay-Delta food web, relatively little is known about the distribution and concentrations and durations of exposure to those toxins, and their trophic ramifications. Additionally, the acute and chronic toxicological effects of cyanobacterial toxins on POD fish species are unknown. Because climate change and human activity in the Bay-Delta ecosystem might increase the intensity and extent of cyanobacterial blooms in the future, more extensive investigations of the dynamics of production, exposure to, and effects of cyanobacterial toxins are warranted. An adaptive approach analogous to that recommended for ammonia/um toxicity testing in Research Topic 12 would be advisable, so limited resources can be more cost-effectively apportioned between laboratory studies and field monitoring.

Research Topic 13 – Field observations of POD organisms:

Because POD organisms are exposed to numerous anthropogenic and "natural" (e.g., cyanobacterial) toxins and other stressors in the Bay-Delta ecosystem, the cause(s) of the POD will be difficult to attribute to ammonia/um unless comparisons of acute and/or chronic toxicity test results to exposure concentrations in the field demonstrably implicate ammonia/um. Therefore, it is not likely that exclusive reliance on laboratory toxicity tests will be productive. Instead, a parallel effort to monitor the physiological status of POD species in and test their responses to Bay-Delta waters is needed.

No systematic historical fish-health data are available for POD fishes, and no systematic monitoring has been proposed to establish a "baseline" to which future fish health can be compared. Routine fish-health protocols are available and are not expensive to implement (Adams et al. 1993). Therefore, such monitoring should be initiated soon.

However, routine fish-health analyses provide only relatively crude indices and are unable to discriminate among numerous chemical stressors. Therefore, priority should be placed on development of genomic arrays that can discriminate the numerous chemical stressors that might be affecting the POD fishes. As a minimum, initial efforts should focus on genomic responses to ammonia/um, metals (e.g., Hg, Se), major-use pesticides upstream and in the Bay-Delta system, and, if possible, toxins produced by *Microcyctis* (i.e., microtoxins). Resident POD fish could be monitored for these genomic responses, and naïve fish could be assayed after *in situ* exposure to Bay-Delta waters (e.g., in flow-through exposure systems on shore or on a floating laboratory). In addition to being useful for monitoring POD species exposed to current stressors, these genomic techniques will be important tools for monitoring the effects of future changes as new stressors emerge and current stressors decline or intensify in the Bay-Delta ecosystem.

Although biomarkers of any type can provide useful information about exposure to toxins and the potential for adverse effects, the interpretation of a positive result in a biomarker assay is challenging because organisms can successfully compensate for many biomarker responses and thus not be affected at the organism or population level. For example, demonstration of a genome-level effect of ammonia/um

(e.g., either an up-regulation or a down-regulation of an enzyme system) in a POD species will not be conclusive evidence that ammonia/um is adversely affecting that population. Therefore, like other biomarkers, genomic arrays will be most useful in ruling-out potential adverse effects of ammonia/um (or other toxins). Additionally, they can add to the weight-of-evidence about a given toxin if a genome-level response is detected. However, a positive genomic biomarker response alone is not sufficient cause-effect evidence for higher-level toxicological responses.

CONCLUSIONS

The Delta-Suisun Bay component of the San Francisco Bay ecosystem is a hydrologically, biogeochemically and trophically complex continuum that is exposed to a large number of anthropogenic and natural stressors, including ammonia/um and other N species. Although numerous studies of individual stressors and ecosystem components have been conducted, are in progress, and are planned, relatively little effort has been devoted to an over-arching analysis of the results and to development of an integrative model of the effects of N and other stressors on POD species and the rest of the Bay-Delta food web.

We strongly recommend that the amount of effort expended on integrative analyses of the data and modeling of the Bay-Delta system be increased substantially. An overarching model would be instrumental in guiding current and future research, integrating the wide variety of information that is currently available and being generated and predicting outcomes of potential management actions and other changes upstream and in the Bay-Delta region. A crucial consideration for developing such a model will be adaptability, so knowledge about underappreciated and yet to be discovered aspects of the current system and emerging stressors can be incorporated into the model framework and structure.

Furthermore, additional field studies of N sources, transformations, and fate are needed, particularly in the Sacramento River portion of the Delta and in Suisun Bay. Results from these empirical studies are needed to parameterize and calibrate the overarching models more accurately, and to help managers make decisions about point and non-point sources of ammonia/um.

Finally, because the Bay-Delta ecosystem is downstream of, surrounded by, and infused with large human populations and/or associated activities, continual change is inevitable. Therefore, the research conducted in and the models of the Bay-Delta system must be flexible, to maintain scientific credibility and be responsive to changing needs of resource managers.

LITERATURE CITED

Adams, S.M., A.M. Brown and R.W. Goede. 1993. A quantitative health assessment index for rapid evaluation of fish condition in the field. *Transactions of the American Fisheries Society* 122:63-73.

Adolf, J.E., C.L. Yeager, M.E. Mallonee, W.D. Miller and L.W. Harding, Jr. 2006. Environmental forcing of phytoplankton floral composition, biomass, and primary productivity in Chesapeake Bay, USA. *Estuarine, Coastal and Shelf Science* 67:108-122.

Cherry, J.A., A.K. Ward and G.M. Ward. 2009. The dynamic nature of land-water interfaces: Changes in structure and productivity along a water depth gradient in the Talladega Wetland Ecosystem. *Verhandlungen International Vereinigung Limnologie* 30(6): in press.

Cloern, JE. 1982. Does the benthos control phytoplankton biomass in south San Francisco Bay? *Marine Ecology Progress Series* 9:191-202.

Cole, B.E., J.K. Thompson and J.E. Cloern. 1992. Measurement of filtration rates by infaunal bivalves in a recirculating flume. *Journal of Marine Research* 113:219-225.

Dugdale, R.C., F.P. Wilkerson, V.E. Hogue and A. Marchi. 2007. The role of ammonium and nitrate in spring bloom development in San Francisco Bay. *Estuarine, Coastal and Shelf Science* 73:17-29.

Fisher, T.R., E.R. Peele, J.A. Ammerman and L.W. Harding. 1992. Nutrient limitation of phytoplankton in Chesapeake Bay. *Marine Ecology Progress Series* 82:51-63.

Hazel, C.R., W. Thomsen and S.J.Meith. 1971. Sensitivity of striped bass and stickleback to ammonia in

relation to temperature and salinity. *California Fish and Game* 57:154-161.
Healey, M. 2008. Science and the Bay-Delta. p. 19-35 in: M. Healey (editor), *The State of Bay-Delta Science: 2008*, CALFED Science Program, Sacramento, California.
Healey, M., M. Dettinger and R. Norgaard. 2008. New perspectives on science and policy in the Bay-Delta. pp. 1-18 in: M. Healey (editor), *The State of Bay-Delta Science: 2008*, CALFED Science Program, Sacramento, California.
Jassby, A. 2008. Phytoplankton in the Upper San Francisco Bay Estuary: Recent biomass trends, their causes and their trophic significance. *San Francisco Estuary and Watershed Science* February 2008:1-24.
Kimmerer, W., L. Brown, S. Culberson, P. Moyle, M. Nobriga and J. Thompson. 2008. Aquatic ecosystems. pp. 73-101 in: M. Healey (editor), *The State of Bay-Delta Science: 2008*, CALFED Science Program, Sacramento, California.
Laursen, A.E. and S.P. Seitzinger. 2002. Measurement of denitrification in rivers: An integrated whole reach approach. *Hydrobiologia* 485:67-81.
Lehman, P.W., G. Boyer, C. Hall, S. Waller and K. Gehrts. 2005. Distribution and toxicity of a new colonial *Microcystis aeruginosa* bloom in the San Francisco Bay Estuary, California. *Hydrobiologia* 541:87-99.
Lehman, P.W., G. Boyer, M. Satchwell and S. Waller. 2008. The influence of environmental conditions on the seasonal variation of *Microcystis* cell density and microcystins concentration in San Francisco Estuary. *Hydrobiologia* 600:187-204.
McCutchan, J.H., J.F. Saunders, A.L. Pribyl and W.M. Lewis. 2003. Open-channel estimation of denitrification. *Limnology and Oceanography Methods* 1:74-81.
Mitsch, W.J. 1977. Water hyacinth (*Eichhornia crassipes*) nutrient uptake and metabolism in a north-central Florida marsh. *Archiv für Hydrobiologie* 81:188-210.
Mitsch, W.J. and J.G. Gosselink. 2000. *Wetlands*. 3rd edition. John Wiley & Sons, New York, New York.
Paerl, H.W. 1983. The effects of salinity on the potential of a blue-green algal (*Microcystis aeruginosa*) bloom in the Neuse River Estuary, NC. N.C. Sea Grant Report. 84 pp.
Paerl, H.W. and J. Huisman. 2008. Blooms like it hot. *Science* 320:57-58.
Paerl, H.W. and J. Huisman. 2009. Climate change: A catalyst for global expansion of harmful cyanobacterial blooms. *Environmental Microbiology Reports* 1:27-37.
Paerl, H.W., K.L. Rossignol, N.S. Hall, B.L. Peierls and M.S. Wetz. 2009. Phytoplankton community indicators of short and long-term ecological change in the anthropogenically and climatically impacted Neuse River Estuary, North Carolina, USA. *Estuaries and Coasts*: in press.
Pinckney, J.L., T.L. Richardson, D.F. Millie and H.W. Paerl. 2001. Application of photopigment biomarkers for quantifying microalgal community composition and in situ growth rates. *Organic Geochemistry* 32:585-595.
Randall, D.J. and T.K.N. Tsui. 2002. Ammonia toxicity in fish. *Marine Pollution Bulletin* 45:17-23.
Rudek, J., H.W. Paerl, M.A. Mallin and P.W. Bates. 1991. Seasonal and hydrological control of phytoplankton nutrient limitation in the lower Neuse River Estuary, North Carolina. *Marine Ecology Progress Series* 75:133-142.
Sellner, K.G. 1997. Physiology, ecology, and toxic properties of marine cyanobacterial blooms. *Limnology and Oceanography* 42:1089-1104
Sommer, T., C. Armor, R. Baxter, R. Breuer, L. Brown, M. Chotkowski, S. Culberson, F. Feyrer, M. Gingras, B. Herbold, W. Kimmerer, A. Mueller-Solger, M. Nobriga and K. Souza. 2007. The collapse of pelagic fishes in the Upper San Francisco Estuary. *Fisheries* 32:270-277.
Thompson, J.K. and F.H. Nichols. 1996. Control of a phytoplankton bloom in San Francisco Bay, California by the filter feeding bivalve *Potamocorbula amurensis* [abstract]. *Proceedings of the Pacific Division of the American Association for the Advancement of Science* 13:98.
USEPA (U.S. Environmental Protection Agency). 2002. Short-term methods for estimating the chronic toxicity of effluents and receiving waters to freshwater organisms. Fourth edition. EPA-821-R-02-

013, USEPA, Washington, DC.

Valdes-Weaver, L.M., M.F. Piehler, J.L. Pinckney, K.E. Howe, K. Rosignol and H.W. Paerl. 2006. Long-term temporal and spatial trends in phytoplankton biomass and class-level taxonomic composition in the hydrologically variable Neuse-Pamlico estuarine continuum, NC, USA. *Limnology and Oceanography* 51:1410-1420.

Vanderploeg, H.A., J.R. Leibig, W.W. Carmichael, M.A. Agy, T.H. Johengen, G.L. Fahnenstiel and T.F. Nalepa. 2001. Zebra mussel (*Dreissena polymorpha*) selective filtration promoted toxic *Microcystis* blooms in Saginaw Bay (Lake Huron) and Lake Erie. *Canadian Journal of Fisheries and Aquatic Sciences* 58:1208-1221.

Ward, A.K. and R.G. Wetzel. 1980. Interactions of light and nitrogen source among planktonic blue-green algae. *Archiv für Hydrobiologie* 90:1-25.

Werner, I., L.A. Deanovic, M. Stillway and D. Markiewicz. 2009. The effects of wastewater treatment effluent-associated contaminants on delta smelt. Draft final report. Aquatic Toxicology Laboratory, School of Veterinary Medicine, University of California, Davis, California.

Wilkerson, F.P., R.C. Dugdale, V.E. Hogue and A. Marchi. 2006. Phytoplankton blooms and nitrogen productivity in San Francisco Bay. *Estuaries and Coasts* 29:401-416.