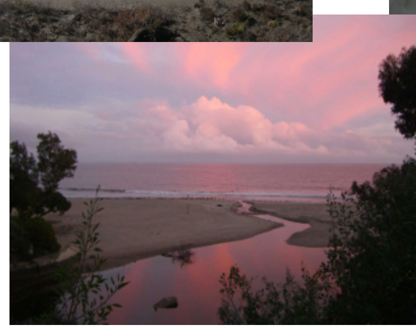
Estuarine Eutrophication

BIGHT'08



Southern California Bight
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Program
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SOUTHERN CALIFORNIA BIGHT 2008 REGIONAL MONITORING PROGRAM: VIII. Estuarine Eutrophication

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Foreword

The Southern California Bight 2008 Regional Monitoring Program (Bight'08) is part of an effort to provide an integrated assessment of environmental condition through cooperative regional-scale monitoring. The Bight'08 program is a continuation of regional surveys conducted in 1994, 1998 and 2003, and represents the joint efforts of more than 90 participating organizations. The Bight'08 program consists of several elements including: Sediment Toxicity, Sediment Chemistry, Areas of Special Biological Significance (ASBS), Demersal Fishes and Megabenthic Invertebrates, Benthic Macrofauna, Offshore Water Quality, Rocky Reefs, Shoreline Microbiology, Bioaccumulation, and most recently, Estuarine Eutrophication. Bight'08 workplans, quality assurance plans, as well as the data described in this report and assessment reports for other elements are available at www.sccwrp.org.

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This report is a product of the dedication and hard work of many individuals who share a common goal of improving our understanding of the environmental quality of the Southern California Bight coastal waters. The authors wish to thank the members of the Bight'08 Estuarine Eutrophication Technical Committee for their assistance with study design, sample analysis, data analysis and report review. This study would not have been possible without the hard work, dedication and exceptional skill of the field sampling team from the following organizations: Tijuana River Estuarine Research Reserve, San Elijo Lagoon Conservancy, Santa Monica Bay Restoration Commission, City of Los Angeles, Natural Resource Conservation Service of the Santa Monica Mountains, California State University Channel Islands, UC Santa Barbara Reserve, and Ventura County. In addition, the Counties of San Diego, Orange, Los Angeles, and Ventura provided sampling support for wet weather sampling to estimate nutrient loads. The authors also wish to express their gratitude to Becky Schaffner (SCCWRP) for assistance with map preparation and Karlene Miller (SCCWRP) for editing this document.

EXECUTIVE SUMMARY

Introduction and Key Questions, and Study Design

The estuaries of the southern California, found in a distinct region that extends from Point Conception to Punta Banda, Baja Mexico, are an important resource for biodiversity, support of commercial and recreational fisheries, migratory birds, endangered species, as well as ecotourism. These estuaries are at risk, due to habitat loss, fragmentation, and increased loading of contaminants from urbanized watersheds. Nutrients are a major form of contaminant loading, particularly from points sources such as industrial and municipal effluent and non-point sources such runoff from agricultural and residential land uses and atmospheric deposition.

While nitrogen (N) and phosphorus (P) are required to support all life forms, too much of a good thing causes problems. Nutrient pollution causes an over-growth of algae and aquatic plants, leading to reduced dissolved oxygen (DO) concentrations, reduced biodiversity and changes in food webs. This collection of symptoms is referred to as "eutrophication." Eutrophication is recognized as one of the leading impairments of water quality in the United States, yet, despite the large number of estuaries in the Southern California Bight (SCB), little data are available on extent of eutrophication and the relationship with watershed nutrient loads. Only three of the SCB's 76 estuaries had sufficient data to be included in the 2007 National Oceanographic and Atmospheric Administration (NOAA) National Estuary Eutrophication Assessment. As the State Water Resources Control Board (SWRCB) prepares to develop estuarine nutrient objectives, a heightened need exists to identify appropriate indicators, standard protocols and methods to interpret data, and establish linkages between nutrient inputs and symptoms of eutrophication to support improved nutrient management.

The Bight 2008 Estuarine Eutrophication Assessment provided an opportunity to conduct the first large scale assessment of estuarine eutrophication in the region, in addition to getting early agreement on indicators and standard protocols, and informing the development of estuarine nutrient objectives. Working together, environmental managers from twenty-one organizations, including stormwater agencies, municipalities, State and Federal regulatory agencies, and scientists joined forces to answer three basic questions:

1) What is the extent and magnitude of eutrophication in SCB estuaries?
2) Is there a difference in eutrophication between different classes of estuaries or by the degree of tidal flushing?
3) Is there a relationship between the symptoms of eutrophication and nutrient inputs?

Study Approach, Design, and Framework Used to Interpret Data

Magnitude of eutrophication was assessed using macroalgal abundance, phytoplankton biomass, and dissolved oxygen (DO), indicators that have scientifically well-vetted linkages to the ecosystem functions and beneficial uses of estuaries. Total N and P loading from the watershed, as well as 19 other water column and sediment physical and chemical parameters were also measured to determine how site-specific factors affect magnitude and extent of eutrophication.

Because eutrophication is highly spatially variable within an estuary, and because this was a regional assessment, we chose to report on targeted index area (segment) within many estuaries to get a broad estimate of extent of eutrophication across the region. A total of 27 segments in 23 estuaries were randomly selected from a comprehensive list of 76 estuaries. For the majority of systems, the segment represents 50 - 100 % of the estuarine area, but for a subset of systems (7 estuaries) the segment represents less than 25% of the total area). Segments were proportionally selected from the list to be able to investigate differences by estuarine classes (enclosed bay, lagoon, river mouth, estuaries) and degree of tidal restriction within the estuary, which relates to tidal inlet status: open, restricted, or closed. Each segment was located in a region of the estuary that is likely to have a longer residence time in order to capture where symptoms of eutrophication would likely be most severe. DO and phytoplankton biomass were assessed continuously while macroalgal biomass and other parameters assessed every other month from November 2008-October 2009.

Reporting on the extent of eutrophication requires a framework to interpret data. Dissolved oxygen, macroalgae, and phytoplankton assessment frameworks for European Union Water Framework Directive (EU-WFD) were applied to the monitoring data set to assess extent of eutrophication. Ecological condition in each segment was classified into one of five categories from very high (minimally disturbed conditions), high, moderate, low, to very low (severely degraded condition) for each indicator.

Study Findings

Question 1: What Is The Extent and Magnitude of Eutrophication in SCB Estuaries?

According to the EU-WFD framework, this study found that eutrophication is pervasive in the SCB segments monitored during the Bight'08 survey. The EU-WFD suggests management action if ecological condition is listed as "moderate" or worse. In SCB estuaries, 78% of segments using macroalgal abundance, 39% using phytoplankton biomass, and 63% using dissolved oxygen were categorized in "moderate" ecological condition or below. Applying a conservative "one out, all out" approach in determining ecological status, wherein the lowest score for any single indicator becomes the overall score the waterbody, all but one of the segments (96%) assessed would require management action. Utilizing a multiple lines of evidence approach that combines the worst of the primary symptoms (phytoplankton or macroalgal biomass) and a secondary symptom (low DO) would result in 53% of segments requiring management action. These findings should be interpreted with caution because segments do not represent the entire estuary and segments from larger estuaries were located within the part of the estuary where we would be most likely to find a problem, if it exists. Thus, these study results cannot be used to infer that the same percentage of estuarine habitat within the SCB is eutrophied. These results represent a preliminary regional estimate of the extent and magnitude of eutrophication in susceptible segments of SCB estuaries, and do not address the spatial extent of eutrophication in any single estuary.

Another use of the eutrophication data set is to provide the ranks of the estuarine segments relative to one another individually for each of the indicators, or overall by integrating across all indicators. Use of the data in this fashion provides context for the severity of eutrophication in the segment relative to other estuaries in our region. The three response indicators did not necessarily score the segment the same, reflecting a difference in dominant algal group (phytoplankton versus macroalgae) or relative importance of direct versus indirect effect of eutrophication (algae versus dissolved oxygen). Generally, macroalgae was the dominant primary producer, though several exceptions were found.

Question 2: Is There a Difference in Eutrophication between Different Classes of Estuaries or by the Degree of Tidal Flushing?

Environmental managers were interested in testing the effect of estuarine class and degree of tidal restriction (i.e., inlet status) on extent of eutrophication. Enclosed bays are the largest, deepest estuaries, and have well-flushed, permanent connections to the ocean. In comparison, the smaller lagoons and river mouth estuaries, which have sand bars that form across their mouths, have intermittent restriction or complete closure of their tidal inlets. Although we hypothesized that the magnitude of eutrophication would be higher in estuaries with more restricted hydrology (longer residence times), we found that class had no effect on extent of eutrophication in the segments studied; nutrient or organic matter loading was more important than inlet status in terms of nutrient impairment. However, macroalgal biomass decreased significantly where tidal variation in water level increased. In addition, the relationship between algal biomass and N and P loads became more significant when the volume and residence time of water in the estuary were both taken into account. Water residence time is largely driven by the status of the ocean inlet and volume is a function of the morphology of the estuary. Furthermore, extent of low DO was significantly related to sediment organic matter, which is typically preserved in habitats that tend to deposit fine-grained sediment in areas of restricted flow. Among paired restricted and unrestricted segments, restricted segments were ranked lower compared to unrestricted segments in the same estuary for nutrient impairment. Thus, inlet status likely influences extent of eutrophication, but is not necessarily more important than gradients in nutrient and organic matter loading.

Question 3: Is There a Relationship between the Symptoms of Eutrophication and Nutrient Inputs?

Eutrophication is assessed based on ecological response indicators, but impairment is managed largely by reducing nutrient inputs. This question sought to determine if relationships exist between nutrient availability and the magnitude of eutrophication symptoms in SCB estuaries, and whether these relationships were stronger for estuarine nutrient concentrations or nutrient loads delivered to the estuary. This question is important for two reasons. First, management strategies differ by whether the ultimate endpoint is defined by nutrient loads to the estuary (weight per unit time) or estuarine water column nutrient concentrations; strategies to control wet weather inputs, representing most of the load, are different from strategies to reduce concentrations, which are more applicable during dry weather. Second, the current USEPA approach to nutrient objectives is driven by the assumption that estuarine nutrient concentrations are a good predictor of eutrophication. We used statistical models to determine the strength of the relationships between extent of eutrophication and nutrient concentrations or nutrient loads.

Watershed nutrient loads and estuarine water-column nutrient concentrations were both significantly, positively correlated with aquatic primary producer (aquatic primary producers) biomass (i.e., macroalgae and phytoplankton), Several important points emerge from the analyses: 1) the relationship between nutrient inputs (water column concentrations and loads) and aquatic primary producer biomass was generally weak, though better for phytoplankton than macroalgae; 2) estuarine water column concentrations had a higher correlation with aquatic primary producer biomass than nutrient loads; 3) selecting the appropriate timescales over which to average the data is important to the strength of the relationship; 4) total nutrients were better correlated with biomass than dissolved inorganic nutrients; and 5) watershed nutrient loads and ambient nutrient concentrations at the segment site were significantly correlated with one another on annual timescales. The relationship between nutrient loads and aquatic primary producer biomass was only significant when estuarine volume and residence time are taken into account. While, these positive relationships build confidence in the use macroalgae and phytoplankton biomass as indicators of eutrophication in SCB estuaries, these models are more indicative of the expression of eutrophication symptoms along a disturbance gradient. Much needs to be done before the models can be used to set site-specific water quality goals to prevent or mitigate eutrophication.

In contrast to algae, extent of low DO events had no significant correlation with N and P loads; instead, it was strongly related to sediment organic matter (OM) content (Section IV). Macroalgae was also significantly correlated with sediment OM. Sediment OM generally increases along a gradient of increasing eutrophication, with increased amounts of OM due to long-term accumulation of external OM loading and/or within-estuary production and accumulation of algae over decadal time-scales (i.e., evidence of past nutrient loading). Thus, DO and algae indicators integrate the effects of increased nutrient loading over very different time-scales. Aquatic primary producer biomass reflects a more immediate response to nutrient loads entering on that particular year, while low DO is largely driven by the combination of OM loading and aquatic primary producer biomass which has accumulated over time. These findings have important implications for how different response indicators can be used for management of eutrophication in estuaries. If nutrient loads are reduced, one may expect to see a response in algal blooms relatively quickly, while hypoxia may decrease over a much longer time.

Recommended Next Steps

Create An Assessment Framework Appropriate For California Estuaries. The State Water Resources Control Board (SWRCB) is in the process of developing nutrient objectives based on ecological response indicators, which will require assessment frameworks specific to the local ecology of our estuaries. Results from this study can be utilized to inform this process by highlighting which indicators are relevant and how sensitive the results are to threshold selection, spatial and temporal sampling, as well as spatial and temporal integration of the data. Furthermore, the experience of our Bight planning committee can be used to refine protocols to optimize monitoring for eutrophication by identifying trade-offs between more data and a better assessment. However, there are still a number of issues that must be addressed to create a scientifically defensible assessment framework for the state of California. These issues include protocol refinement, science supporting the selection of thresholds, determination of how to incorporate inter-annual and spatial variability, how to incorporate duration (length and frequency) of blooms and hypoxia, and recognition that eutrophication may occur naturally in some of the smaller seasonally closed estuaries.

Refine Predictive Load - Response Models. Analysis of the relationships between nutrient loading and ecological response in this study was limited to simple statistical models. While a relationship between algae and nutrient loads was significant, these models lacked precision and are not yet appropriate for management use. The predictive capability of these models can be improved through: 1) development of, at minimum, improved data and models of estuarine hydrology, shown to be critical in improving load-response relationships, and 2) mechanistic studies of processes known to mitigate the effects of eutrophication (e.g., denitrification, etc.). This information can be incorporated into a regional model for scenario analysis of various nutrient loading rates and expected estuarine response.

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I. INTRODUCTION

Purpose of Document

The Southern California Bight (SCB; Figure I-1) is an open embayment in the coast between Point Conception, California and Cabo Colnett (south of Ensenada), Baja California. Complex bathymetry and currents have resulted in a diversity of habitats and marine organisms, including more than 500 species of fish and several thousand species of invertebrates. The SCB is a major migration route for marine bird and mammal populations and is ranked among the most diverse ecosystems in northern temperate waters. In addition to its ecological value, the coastal zone of the SCB is a substantial economic resource. The SCB is home to more than 20 million people (NRC 1990) and southern California receives over 100 million visitors to its beaches and coastal areas annually. The combination of resident and transient population has resulted in highly developed urban environment that has greatly altered the natural landscape. The conversion of open land into impervious surfaces has included dredging and filling over 75% of bays and estuaries and extensive alterations of coastal streams and rivers (Brownlie and Taylor 1981, Horn and Allen 1985, NRC 1990, Zedler 1996). This “hardening of the coast” changes both the timing and rate of runoff releases to estuaries coastal waters and can affect water quality through addition of sediment, toxic chemicals, pathogens, and nutrients.

Figure I-1. Study area.

Although the effects of these anthropogenic stressors to the SCB are not well understood, in all cases a Bight-wide perspective is needed to coordinate, contextualize and manage these effects. The Southern California Bight (SCB) Regional Monitoring Program is an integrated, multi-disciplinary and multi-institutional study that provides a unique platform for collecting data for Bight-wide perspectives. The SCB Regional Monitoring Program is a partnership of more than 60 organizations collaborating to address management questions of regional importance in the Bight offshore, nearshore and estuarine habitats. The Bight surveys provide a mechanism to develop standardized methods, quality assurance protocols and data transfer standards agreeable to all participants. This ensures that all data collected during the survey can be integrated and provides the foundation for enhanced coordination among southern California's monitoring programs. The surveys have also provided a forum for multi-party agreement about ways to analyze and interpret marine and estuarine monitoring data. "Core" components of Bight surveys include: 1) offshore water quality, 2) coastal ecology, focusing on sediment quality, and 3) shoreline microbiology. Estuarine eutrophication is a new study component of the SCB Regional Monitoring Program, conducted during the Bight 2008 reporting cycling.

The purpose of this document The Southern California Bight 2008 Regional Monitoring Program Report provides a summary of the findings, recommendations and conclusions from the Estuarine Eutrophication Assessment of the SCB Regional Monitoring Program.

Background and Context

Eutrophication is the increased production of organic matter from aquatic algae and plants. Cultural eutrophication of estuaries and coastal waters is a global environmental issue, with demonstrated links between anthropogenic changes in watersheds, increased nutrient loading to coastal waters, harmful algal blooms, hypoxia, and impacts on aquatic food webs (Valiela *et al.* 1992, 1997; Zaldivar *et al.* 2008). These ecological impacts of eutrophication of coastal areas can have far-reaching consequences, including fish-kills and lowered fishery production (Glasgow and Burkholder 2000), loss or degradation of seagrass and kelp beds (Twilley 1985, Burkholder *et al.* 1992, McGlathery 2001), smothering of bivalves and other benthic organisms (Rabalais and Harper 1992), nuisance odors, and impacts on human and marine mammal health from increased frequency and extent of harmful algal blooms and poor water quality (Bates *et al.* 1989, 1991; Trainer *et al.* 2002). These modifications have significant economic and social costs.

In California, the impacts of nutrient loading on estuaries and coastal waters have not been well monitored, with the notable exception of San Francisco Bay (Cloern 1989, 1996). In southern California, only two of the region's 76 estuaries were included in the National Oceanographic and Atmospheric Administration's (NOAA) National Estuarine Eutrophication Assessment Report (Bricker *et al.* 1999). The need to monitor the magnitude of eutrophication in southern California estuaries is heightened by the State Water Resources Control Board's intent to develop nutrient objectives for estuaries. Data from southern California Bight estuaries would help to drive the selection of appropriate indicators, shed light on critical conditions for assessment with those indicators, and provide context for discussion of thresholds.

Goal and Objectives of Estuarine Eutrophication Assessment

The Bight 2008 Estuarine Eutrophication Assessment provided an opportunity to conduct the first large scale assessment of estuarine eutrophication in the region, get early agreement on indicators and standard protocols, and inform the development of estuarine nutrient objectives. Working together, environmental managers from twenty-one organizations, including stormwater agencies, municipalities, State and Federal regulatory agencies, and scientists joined forces to answer three basic questions:

1) What is the extent and magnitude of eutrophication in SCB estuaries?
2) Is there a difference in eutrophication between different classes of estuaries or by the degree of tidal flushing?
3) Is there a relationship between the symptoms of eutrophication and nutrient inputs?

The first question seeks to document the magnitude and extent of eutrophication in SCB estuaries. The answer to this question should reflect magnitude (how much of a problem) and extent (the duration, e.g., chronic or episodic as well as spatially). As a regional survey, the emphasis was on assessing as many estuaries as possible, rather than a complete spatial characterization of a handful of estuaries.

The second question seeks to evaluate the differences between three estuarine classes: enclosed bays, lagoons and river mouth estuaries. Estuaries within southern California are highly variable in how they respond to nutrient loading due to differences in tidal forcing, freshwater residence time, salinity regime, stratification, denitrification, and other factors. This combination of factors results in differences in the dominant aquatic primary producer communities (i.e., phytoplankton, macroalgae, submerged aquatic vegetation, etc.).

The third question seeks to determine whether a dose-response relationship exists between nutrient inputs and estuarine biological response. Total nitrogen and phosphorus loads into each estuary are being estimated as a component of the Bight'08 Offshore Water Quality study. The approach being used to develop annual loads will provide a coarse estimate. The eutrophication assessment will use these data, in an exploratory fashion, with the intent to establish whether a dose-response relationship exists over a gradient of disturbance captured by these estuaries.

Two special studies were conducted in conjunction with the eutrophication assessment. The first seeks to assess the presence of cyanobacteria in fresh and brackish coastal habitats. The second will use stable isotopes of nitrogen and oxygen to assess nitrogen sources and cycling within three of the 23 estuaries being sampled.

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II. EXTENT AND MAGNITUDE OF EUTROPHICATION IN SOUTHERN CALIFORNIA ESTUARIES

Introduction

Eutrophication of estuaries is a global environmental issue, with demonstrated links between anthropogenic changes in watersheds, increased nutrient loading to coastal waters, harmful algal blooms, hypoxia, and impacts on aquatic food webs (Valiela *et al.* 1992, Smith *et al.* 1999, Kamer and Stein 2003). These ecological impacts of eutrophication of coastal areas can have far-reaching consequences, including fish-kills and lowered fishery production (Glasgow and Burkholder 2000), loss or degradation of seagrass and kelp beds (Twilley 1985a, Burkholder *et al.* 1992, McGlathery 2001), smothering of bivalves and other benthic organisms (Rabalais and Harper 1992), nuisance odors, and impacts on human and marine mammal health from increased frequency and extent of harmful algal blooms and poor water quality (Bates *et al.* 1989, 1991; Trainer *et al.* 2002). These modifications have significant economic and social costs (Turner *et al.* 1998). According to the United States Environmental Protection Agency (USEPA), eutrophication is one of the top three leading causes of impairments of the nation's waters (USEPA 2001). Scientifically-based methods to diagnose eutrophication as well as surveys to document the extent and magnitude of the problem are needed as the foundation for protecting pristine systems, identifying adverse effects, and providing targets for restoration or mitigation of systems where adverse effects of eutrophication have already occurred.

Over the past decade, much work has been done to establish standardized methodologies to assess eutrophication (Bricker *et al.* 2003, Zaldivar *et al.* 2008) and conduct surveys to evaluate the magnitude and extent (Bricker *et al.* 1999, Borja *et al.* 2009a, Devlin *et al.* 2011, Garmendia *et al.* 2012). Most recently, a 2007 study of US estuaries conducted through the NOAA National Estuarine Eutrophication Assessment (NEAA) found that the majority of estuaries assessed had overall eutrophic conditions rated as moderate to high and that the most commonly occurring eutrophic symptom was high spatial coverage and frequency of elevated chlorophyll a levels, although most estuaries also exhibited at least one other moderate to high symptom (e.g., dissolved oxygen). Furthermore, on the whole, eutrophication in US estuaries remained largely unchanged from 1999 to 2004 and survey participants expected most systems to worsen by 2020 (Bricker *et al.* 1999). The survey also served to highlight data gaps. For example, in the Southern California Bight (SCB), one of the most populated regions in the US, sufficient data was available to make an assessment in 2 estuaries out of the region's 76 total. Among SCB estuaries, protected embayments, which represent 78% of the areal extent of estuarine habitat but 17 % by number, tend to be most data rich (e.g., (Boyle *et al.* 2004, Nezlin *et al.* 2007, Nezlin *et al.* 2009)). In comparison, the smaller "bar-built" lagoons and river mouth estuaries, which represent 22% of the areal extent but 82% by number, are tremendously data poor (Fong and Zedler 2000). These "bar-built" estuaries, typical of Mediterranean climates (Largier *et al.* 1997), are so called because they have sand bars form across their mouths, often resulting in the intermittent restriction or complete closure of these estuaries to surface water tidal exchange (Webb *et al.* 1991, Largier *et al.* 1996). These intermittently tidal estuaries are known globally for their increased susceptibility to eutrophication during their "closed" state due to restricted flushing (Painting *et al.* 2007, Zaldivar *et al.* 2008), but these are the estuaries in Southern California that are the least well characterized with respect to eutrophication.

The SCB Regional Monitoring Program (RMP) provided an opportunity to characterize the ecology and status of eutrophication in SCB estuaries. The SCB RMP is an integrated, multi-disciplinary and multi-institutional program that provides a unique platform for collecting data for Bight-wide perspectives on a number of management questions of regional importance. We conducted an assessment of eutrophication in 27 segments in 23 estuaries from Point Conception, California and Cabo Colnett (south of Ensenada), Baja California from November 2008 through October 2009. This study aimed to address three questions: 1) What is the magnitude and extent of eutrophication in SCB estuaries? 2) Is there a difference in eutrophication between different classes of estuaries or by the degree of tidal flushing? 3) Is there a relationship between the symptoms of eutrophication and nutrient inputs? It should also be noted that the objective of the study is to estimate extent and magnitude of eutrophication in SCB estuaries regionally and is not meant to be an exhaustive analysis of any single estuary.

In this section, we report on the results of the assessment of the magnitude and extent of eutrophication. Companion sections (Sections IV and V) report on the analysis of the relationship between nutrient loads and the expression of eutrophication in SCB estuaries. In addition, we used this survey to explore the applicability of two existing eutrophication assessment framework to southern California estuaries, with the intent to understand how assessment approach and data integration affect the results of the assessment.

Review of Existing Assessment Frameworks for Application to SCB Estuaries

Several studies have demonstrated the shortcomings of using estuarine nutrient concentrations or loads alone to predict eutrophication (Cloern 2001, Kennison *et al.* 2003, Devlin *et al.* 2007). Estuaries are highly variable in how they respond to nutrient loading due to differences in physiographic setting, salinity regime, frequency and timing of freshwater flows, magnitude of tidal forcing, sediment load, stratification, residence time, denitrification, and other factors (Dettmann 2001, Pinckney *et al.* 2001, Zaldivar *et al.* 2008, Duarte 2009, Duarte *et al.* 2009). This combination of “co-factors” results in differences in the dominant primary producer communities (i.e., phytoplankton, macroalgae, benthic algae, submerged aquatic vegetation, emergent macrophytes). It can also alter the way nutrients are cycled (taken-up, stored, and remineralized) within the estuary. At times, these co-factors can play a larger role in mitigating estuarine response to nutrient loads or concentrations, blurring or completely obscuring simple nutrient limitation of primary production (Peckol *et al.* 1994, Pinckney *et al.* 2001, Duarte *et al.* 2009). The analysis of Bight Eutrophication Assessment data shows a weak correlation between water column nutrient concentrations and aquatic primary producer biomass and no correlation with dissolved oxygen. This is understandable given that biological communities in the water column and sediments continuously uptake and recycle nutrients, potentially reducing surface water concentrations to non-detectable levels (Cloern 1996, 2001). Because of the unreliability of nutrient concentrations or loads in diagnosing eutrophication, there has been a shift towards the use of ecological response indicators to estimate extent and magnitude of eutrophication (e.g., algal abundance, dissolved oxygen; (Bricker *et al.* 2003, Devlin *et al.* 2007, Zaldivar *et al.* 2008)).

A number of assessment frameworks have been developed to assess the status of eutrophication in estuaries (Bricker *et al.* 2003, Devlin *et al.* 2007, Zaldivar *et al.* 2008). The most representative assessment frameworks incorporate annual data with sampling throughout the year, inclusion of frequency of occurrence and spatial extent in indicator metrics, and use of a combination of indicators into an overall condition rating (Devlin *et al.* 2011). We chose to apply two assessment frameworks to the Bight dataset: 1) the Assessment of Estuarine Trophic Status (ASSETS), and 2) Single Indicator frameworks proposed for use as a part of the European Union Water Framework Directive (EU-WFD).

Briefly, ASSETS was developed to assess the status of eutrophication in US estuaries through NOAA's National Estuarine Eutrophication Assessment (NEEA). It is an integrative and adaptive assessment method that assesses overall trophic status of an estuary using an assessment of pressure (Overall Human Influence- OHI: nutrient loads and susceptibility of the system/flushing rate), state (Overall Eutrophic Condition- OEC: evaluation of biological response variables), and expected response (Definition of Future Outlook- DFO: prediction of future pressures from the watershed) (Bricker *et al.* 2003, Whitall *et al.* 2007a, Bricker *et al.* 2008). The approach uses a combination of primary and secondary indicators to derive an OEC index which is combined with a measure of OHI and the DFO to provide a single grade for classifying estuarine systems into one of five categories. Primary symptoms of eutrophication include: chlorophyll a (concentration, spatial coverage, and frequency), and epiphyte and macroalgae (best professional judgment assessment of problem, and frequency). Secondary symptoms include: dissolved oxygen (concentration, spatial extent, frequency), loss of submerged aquatic vegetation (heuristic assessment of problem, magnitude of loss), and nuisance and toxic blooms (heuristic assessment of problem, duration, frequency). Because the OEC index is the most developed component of the ASSETS framework, comparisons between estuaries tend to be based on state, though ASSETS is currently being refined to better clarify the link between pressure, state and response so that the framework can be used to encourage more proactive approaches to maintenance of estuarine health (Bricker *et al.* 2003). Furthermore, the authors of ASSETS aim to contribute to the EU-WFD classification system to develop a unified approach for assessing eutrophication in estuaries and coastal waters.

The EU-WFD was developed as a means of regulating and monitoring water bodies in European Union member states by focusing on the overall ecology and function of ecosystems, organizing management of waterbodies by catchment and standardizing protocols across Europe (Borja *et al.* 2006, Hering *et al.* 2010). However, it has only an implicit requirement to assess eutrophication, requiring an evaluation of a suite of biological, physio-chemical and hydro-morphological quality elements that together determine the ecological status of a water body with respect to nutrients as well as other pollutants (Borja *et al.* 2006). It defines deterioration and improvement of "ecological quality" in a water body based on biological response indicators, rather than by changes in ambient physical or chemical variables alone. An overall assessment of each water bodies takes into account human driving forces (watershed characteristics like population), nutrient pressure (loading and dilution potential), state of the ecosystem (changes in biogeochemical processes),impact based on ecological quality elements (divergence of biological response variables from reference condition) and response (likely management actions to reduce impacts; (Borja and Dauer 2008, Borja *et al.* 2011)). The objective of the EU-WFD is to achieve at least a "high ecological quality status" for all EU waterbodies by 2015 (Borja *et al.* 2006).

Because the Bight'08 Eutrophication Assessment was a one year synoptic study of estuarine condition and because the watersheds and estuaries were not comprehensively characterized for pressure and susceptibility factors, we could not conduct an overall assessment of ecological condition for each system. Instead, we extracted the biological response indicators from each framework for an indicator-specific analysis of extent and magnitude of eutrophication. Of the available frameworks, the EU-WFD lent itself to this process somewhat easier than ASSETS, primarily because indicators are separated into individual "elements" with numeric thresholds for classifying ecological condition into one of five categories for each indicator. ASSETS uses an integrated multi-metric approach, and some indicators have numeric thresholds (e.g., phytoplankton, dissolved oxygen) and other indicators are classified as "problem/no problem" (e.g., macroalgae). Ultimately, we chose to use the EU-WFD to make an assessment of extent and magnitude of eutrophication in southern California Bight estuaries for three indicators (dissolved oxygen, phytoplankton and macroalgae) because 1) dissolved oxygen thresholds used by the EU WFD are more closely aligned with a recent review of dissolved oxygen criteria for California estuaries (Sutula *et al*. 2012) and 2) the EU WFD has numeric thresholds for macroalgae, a dominant primary producer in SCB estuaries (Section III). However, we used the two frameworks to conduct a sensitivity analysis on how approach and differences in data-integration results.

Methods

Study Area

The Southern California Bight (SCB; Figure II-1) is an open embayment in the coast between Point Conception, California and Cabo Colnett (south of Ensenada), Baja California. The SCB is host to 76 estuaries ranging in size from 1 ha to over 50,000 ha and is home to a diversity of organisms, including more than 500 species of fish and several thousand species of invertebrates. The SCB is a major migration route for marine bird and mammal populations and is ranked among the most diverse ecosystems in north temperate waters. In addition to its ecological value, the coastal zone of the SCB is a substantial economic resource. The SCB is home to more than 20 million people (NRC 1990) and southern California receives over 100 million visitors to its beaches and coastal areas annually. The combination of resident and transient population has resulted in highly developed urban environment that has greatly altered the natural landscape. The conversion of open land into impervious surfaces has included dredging and filling over 75% of bays and estuaries and extensive alterations of coastal streams and rivers (Brownlie and Taylor 1981, Horn and Allen 1985, NRC 1990, Zedler 1996). These changes to hydrology alter both the timing and rate of runoff releases to coastal waters and can affect water quality through addition of sediment, toxic chemicals, pathogens, and nutrients. The coastal watersheds that drain to the SCB are comprised of approximately 14,000 km^2 of urban and agricultural land uses (Ackerman and Schiff 2003).

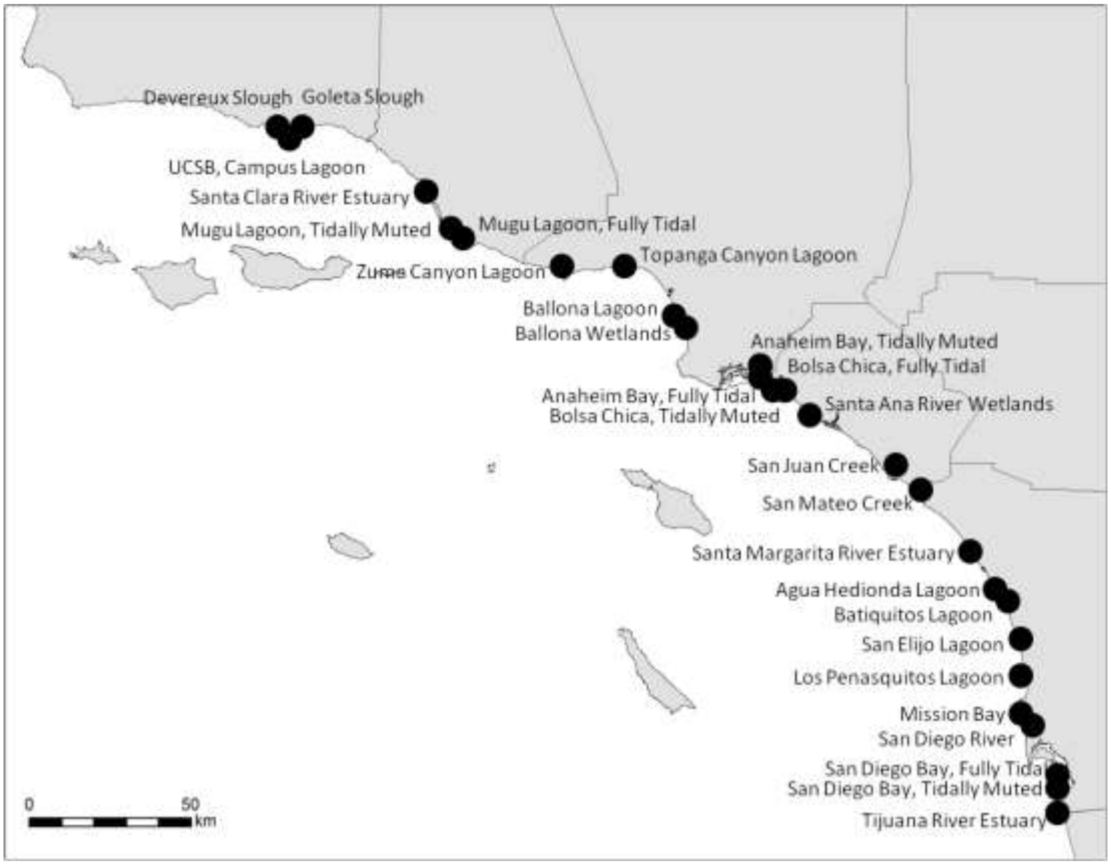


Figure II-1. Map of Bight'08 **Estuaries Eutrophication Assessment segment sites.**

The SCB has a Mediterranean climate, with an average annual rainfall of 10–100 cm (e.g., Nezlin and Stein 2005), falling primarily during winter months (December through March), and approximately 20 annual storm events (Ackerman and Weisberg 2003). Winter runoff to the SCB contributes more than 95% of the total annual runoff volume (Schiff *et al.* 2000, Ackerman and Weisberg 2003). However, urban runoff through the year contributes substantial loads of contaminants to SCB estuaries (Sengupta *et al.* submitted).

Site Selection

The Bight'08 Eutrophication Assessment study design was a probability-based survey in which sites were randomly selected from a comprehensive list of 57 estuaries (excluding ports, marinas, and estuaries undergoing restoration at the time of the survey). Survey design takes into account the three subpopulations of interest to Bight'08 participants: 1) Estuarine class (enclosed bays, lagoons, and river mouth estuaries), 2) Tidal regime (perennially, intermittently, and ephemerally open to surface water tidal exchange), and 3) Presence or absence of anthropogenic muting of tidal regime, defined by 100% containment of the segment by dikes, levees and/or weirs that reduce the amplitude of tidally-induced water level fluctuations in the estuary. While not sampled as separate strata in the survey, some weighting took place to emphasize sampling of selected classes and tidal regimes (Table II-1).

Each estuary within the study region was assigned one of three classes (geoform): enclosed bay, lagoon or river mouth. The system was also attributed tidal inlet status describing the frequency of surface water tidal exchange (open, diked, closed). Estuaries that had sub-regions that were fully tidal and had a section that was anthropogenically muted (diked) were entered twice in the list of estuaries. Some estuaries were excluded from the sample frame. Small creek mouths less than 10 m in width at the mouth, were felt to have insufficient area for assessment purposes. Open embayments, were felt to be driven by physical factors compared to enclosed bays, river mouths, and lagoons and were also eliminated to ensure comparability among sites in the study. Ports and marinas were excluded from the frame because of additional anthropogenic effects on these systems may interfere with expression of eutrophication. Estuaries currently undergoing restoration efforts were also excluded because restoration efforts (dredging, planting, etc.) may mask expression of eutrophication. Estuaries were selected proportional to the total number of estuaries in each class for enclosed bays and lagoons in the Northern portion of the SCB region (Newport Bay and north). Interest and participation allowed for an intensification of effort in the San Diego Region (Dana Point/San Juan Creek and south); thus all estuaries were selected in the San Diego Region to complete a census of estuaries.

Because this was the first such regional assessment, there was a conscious decision to emphasize the collection of data at index areas across many estuaries, rather than to characterize eutrophication spatially within an estuary. Because eutrophication is highly spatially variable within an estuary, we endeavored to make the results comparable across estuaries by selecting an index area, otherwise referred to as a "segment," which varies in size depending on the estuary (Table II-1). Because the segment in many cases does not represent the entire estuary, reporting on eutrophication will be on a "percent of estuarine segments." The study results represent a conservative estimate of extent and magnitude of eutrophication in SCB estuaries regionally and are not meant to be an exhaustive analysis of any single estuary. The relative percentage of the estuary represented by the segment is listed in Table II-1. Once an estuary was selected for assessment, the segment was selected using the following criteria: 1) segment must be safely accessible throughout the year (this excluded sites used by rare and endangered species for all or part of the year), 2) water residence time was likely to be longest, and 3) public access to segment was limited/restricted. A total of 27 segments were selected in 23 estuaries (Figure II-1; Table II-1). Field and Laboratory Methods

The Bight'08 assessment was a synoptic study of ecological response indicators monitored for one water year. In each of these segments, the magnitude of eutrophication of Southern California estuaries was assessed via a series of ecological response indicators collected during November 2008-October 2009. These biological response indicators have a direct linkage to estuarine ecosystem services and beneficial uses: dissolved oxygen and primary producer abundance. Sampling of primary producer abundance included macroalgal biomass and percent cover, surface water phytoplankton biomass (chlorophyll *a*), benthic algal biomass (sediment chlorophyll *a*), brackish submerged aquatic vegetation (SAV) density and percent cover. Table II-2 gives a detailed list of indicators and analytes measured, the data collection method, and the frequency of measurement.

Table II- 1. Bight'08 **Eutrophication Assessment estuary segments.**

Geoform	Tidal Connection	Name (Code)*	Watershed Area (km^2)	Estuary Area (m^2)	% Estuary in Segment	Habitat Type: Subtidal Unveg	Habitat Type: Subtidal Eelgrass	Habitat Type: Mudflat	Habitat Type: Marsh	Avg Annual Temp (^oC)	Avg Annual Salinity (ppt)	Annual TP Load (kg)	Annual TN Load (kg)
Enclosed Bay	Perennial	Agua Hedionda Lagoon (AHL)	546	1,410,480	50%	60%	17%	11%	12%	19.7	33.2	3,283	57,185
		Batiquitos Lagoon (BQL)	131	2,022,063	50%	18%	29%	20%	33%	18.9	33.1	46,966	60,037
		Bolsa Chica-Full (BCF)	25	1,708,457	50%	62%	8%	21%	10%	20.6	34.4	25	2,500
		Bolsa Chica-Muted (BCM)	59	651,724	50%	72%	0%	7%	20%	21.0	34.6	1,081	5,840
		Mission Bay-Full (MB)	118	8,795,281	15%	18%	75%	5%	2%	23.1	35.2	7,902	41,283
		San Diego Bay-Full (SDF)	1037	50,094,111	10%	63%	22%	13%	2%	20.0	34.2	8,045	147,537
		San Diego Bay-Muted (SDM)	362	17,898,631	5%	28%	0%	71%	1%	19.9	46.9	8,045	147,537
		Anaheim Bay/SealBeach-Full (SBF)	130	4,194,998	50%	28%	11%	5%	57%	20.1	34.3	550	13,832
		Anaheim Bay/SealBeach-Muted (SBM)			10%	28%	11%	5%	57%	21.5	34.9	550	13,832
Lagoon	Ephemeral	San Mateo Lagoon (SMC)	346	126,262	100%	20%	0%	0%	80%	17.7	0.6	656	8,895
		UCSB Campus Lagoon (UCL)	10	124,229	75%	96%	0%	0%	4%	20.1	34.7	222	1,257
	Intermittent	Devereaux Lagoon (DL)	10	231,885	100%	41%	0%	27%	32%	20.7	30.7	3,198	7,287
		Los Penasquitos Lagoon (LPL)	244	1,306,657	50%	7%	0%	7%	85%	18.4	32.7	5,669	28,325
		San Elijo Lagoon (SEL)	210	1,261,824	75%	9%	0%	54%	38%	19.0	24.5	2,080	63,262
	Perennial	Ballona Lagoon -Muted (BL)	354	22,950	100%	71%	0%	29%	0%	20.9	33.7	13,939	95,098
		Ballona Wetlands-Muted (BW)	354	627,857	100%	0%	0%	19%	81%	19.1	30.6	13,939	95,098
		Goleta Slough (GS)	119	804,812	50%	20%	0%	12%	68%	19.7	29.7	8,518	20,369
		Mugu Lagoon-Full (MLF)	803	14,098,340	15%	8%	0%	13%	79%	17.2	23.9	65,281	209,210
		Mugu Lagoon-Muted (MLM)			25%	8%	0%	13%	79%	19.1	34.4	65,281	209,210
		Santa Ana R. Wetlands-Muted (SAR)	4336	331,670	75%	37%	0%	14%	50%	19.4	34.1	16,528	89,981
		Tijuana River Estuary (TJE)	4452	2,755,819	25%	7%	0%	17%	76%	18.0	31.9	82,988	348,018
River Mouth	Intermittent	San Juan Creek (SJC)	458	64,839	100%	35%	0%	65%	0%	20.9	12.7	7,695	30,062
		Santa Clara River (SCR)	4210	1,412,587	75%	50%	0%	48%	2%	20.6	4.9	22,483	239,378
		Santa Margarita Estuary (SME)	1918	1,020,806	75%	33%	0%	24%	43%	19.8	25.3	19,177	137,799
		Topanga Canyon Lagoon (TC)	51	3,678	100%	100%	0%	0%	0%	19.6	17.7	25	187
		Zuma Canyon Lagoon (ZC)	23	17,544	100%	56%	0%	13%	31%	19.9	2.6	154	578
	Perennial	San Diego River (SDR)	1120	1,142,328	50%	45%	0%	23%	32%	20.6	28.2	4,629	54,066

*Sites indicated as "muted" have an anthropogenically muted tidal regime through presence of dikes, tide gates or weirs.

Table II-2. List of indicators measured in the SCB Estuarine Eutrophication Assessment.

Parameter	**Data Collection Method and Location**	**Analytes Measured**	**Frequency**
Dissolved Oxygen	In Situ Data Sonde in bottom water at single location within segment	• Dissolved oxygen percent saturation	Continuous January through October 2009
Macroalgae	3 Transects within designated segment	• Wet and dry weight • Biomass • Taxonomic composition • Percent cover	Every other month November 2008- October 2009
Brackish Water Submerged Aquatic Vegetation	3 Transects within designated segment	• Wet and dry weight • Biomass • Genus • Percent cover	Every other month November 2008- October 2009
Phytoplankton Biomass	Discrete water sample 10 cm below surface in water column at 1 macroalgal transect	• Chlorophyll a and pheopigments	Every other month November 2008- October 2009
	In Situ Data Sonde in bottom water at single location within segment	• Chlorophyll fluorescence	Continuous January through October 2009
Benthic Microalgae	3 composites from surface sediments (0-1 cm) at ends of each macroalgal transect	• Chlorophyll *a* and pheopigments	Every other month November 2008- October 2009
Sediment Quality	3 composites from surface sediments (0-1 cm) at ends of each macroalgal transect	• Percent solids • Sediment TN, TP, and TOC • Grain size	Every other month November 2008- October 2009
Estuarine Nutrient Concentrations	Discrete water sample 10 cm below surface in water column at 1 macroalgal transect	• Water column TN, TP, TDN, TDP, nitrate, ammonium, phosphate, silicate	Every other month November 2008- October 2009
Nutrient Loads	Field sampling of nutrient concentration and continuous flow modeling at watershed mass loading station	• Water column TN, TP, TDN, TDP, nitrate, ammonium, phosphate	Event-based sampling for wet weather, every other month November 2008- October 2009 for dry weather TN and TP; August 2009 for TN, TP and all nutrient forms
Water column Physiochemistry	In Situ Data Sonde in bottom water at single location within segment	• Temperature • Salinity • pH • Turbidity	Continuous January through October 2009

Macroalgae Abundance. Monitoring of macroalgal abundance provided information on when algal blooms occur in each class of estuary, how far they extend spatially, and how long they endure. Macroalgal abundance was determined by measuring percent cover and algal biomass. Within each index area, three 30 - 50 m transects were laid out in the intertidal area, parallel to the water's edge and along the same elevational contour at approximately three quarters of the distance from the mean lowest low water line to the downslope end of vascular vegetation on the mid-to-upper mudflat. This area has been demonstrated to be representative of macroalgae accumulation in southern California estuaries (Kennison *et al.* 2003). Percent cover was measured at ten randomly chosen points along each transect by placing a 0.5 m^2 quadrat with 49 intercepts on the benthos and recording the presence or absence of each macroalgae species under each intercept. Biomass was collected at 5 of the quadrat locations. Each biomass sample was refrigerated until analysis and processed within 24 hours of collection. In the laboratory, algal samples were cleaned of macroscopic debris, mud and animals, and sorted to genus level. Excess water was shed from each sample, which was then weighed wet and dried at 60°C to a constant weight, then weighed dry. During data analysis, all macroalgae genus weights were summed for each quadrat to give a total macroalgae wet and dry weight in each quadrat. Because of lack of confidence in taxonomic expertise among the field groups, a decision was made to lump macroalgal biomass and cover data into broad taxonomic groups (green, red), maintaining wrack separately.

Phytoplankton Biomass. Phytoplankton biomass was estimated from fluorescence measurements collected via *in situ* optical probe (YSI 6600 sonde, chlorophyll fluorescence probe), with bi-monthly discrete chlorophyll *a* water samples taken to calibrate the continuous fluorometry. Discrete suspended chlorophyll *a* pigments were concentrated from 250-500 ml of sample water by filtering at low vacuum through a 45 mm diameter Whatman glass fiber filter. Filters were stored in a petri-dish covered in aluminum foil and frozen until analysis. Photosynthetic pigments were extracted from filters in 90% acetone solution and allowed to steep overnight, to ensure complete extraction of chlorophyll *a* (EPA 445). Fluorescence was measured before and after acidification with 0.1 M HCl to determine the phaeophytin-corrected chlorophyll *a*. Concentrations were calculated relative to a laboratory standard. *In situ* chlorophyll fluorescence was measured every 15 minutes using an optical probe mounted to a YSI 6600 V2 data sonde. Probes were maintained according to factory specifications and were routinely calibrated. Fluorescence measurements were calibrated to chlorophyll *a* concentrations using least-squares regression generated from daily averaged data probe measurements and discrete concentration data collected on that same day.

Microphytobenthos. Benthic chlorophyll *a* (microphytobenthos) was determined on sediment composite samples collected at the beginning and end of each macroalgae transect. Composites were comprised of ten sediment plugs (3 cm in diameter, 1 cm deep) collected at each end of the transect (20 total). The samples were collected downslope of the end of each transect in approximately 30 cm of water depth. Plugs were homogenized in sample bags and refrigerated until analysis. A subsample from each bag was collected in a 15 ml centrifuge tube and frozen for benthic chlorophyll *a* analysis. Benthic chlorophyll *a* samples were analyzed for chlorophyll *a* and phaeopigments as described above for phytoplankton.

***Ruppia spp.* Abundance.** Monitoring of *Ruppia spp.* (brackish water submerged aquatic vegetation) abundance provided information on when and where stands occur in each class of estuary, how far they extend spatially, and how long they endure. *Ruppia spp.* abundance was determined by measuring biomass. Within the subset of estuaries where *Ruppia spp.* was observed in each index area, three transects were laid out perpendicular to the shore-line bisecting the channel. Biomass was collected using sampling "tongs" (Rodusky *et al.* 2005), comprehensively collecting all biomass in a defined area, at 3 to 5 across the channel (at the thalweg, and one or two evenly spaced locations between the thalweg and the water's edge). Each biomass sample was refrigerated until analysis and processed within 24 hours of collection. In the laboratory, biomass samples were cleaned of macroscopic debris, mud and animals, and sorted to genus level. Excess water was shed from each sample, which was then weighed wet, and dried at 60°C to a constant weight, then weighed dry.

Dissolved Oxygen and Water Column Physiochemistry. Water column physiochemistry was measured continuously using a YSI 6600 data sonde. Each sonde was outfitted with a conductivity/temperature sensor, ROX optical dissolved oxygen probe, extended deployment pH probe, chlorophyll optical sensor, and a turbidity optical sensor. All sensors were treated with anti-fouling tape and calibrated at a minimum of once monthly. Sondes were deployed at one location in each segment, in bottom water (approximately 30 cm from the sediment surface). Measurements were collected every 15 minutes throughout the deployment period. Dissolved oxygen concentrations were calculated from percent saturation, temperature and salinity data. An hourly running average was applied to the data set to smooth high frequency noise.

Ambient Nutrients. A single grab sample was collected and subsampled for ambient nutrients (nitrate, nitrite, ammonium, soluble reactive phosphate, total nitrogen, total phosphorus, total dissolved nitrogen and total dissolved phosphorus). A 1 liter amber bottle which was triple rinsed with sample water before filling completely. The sample bottle was open and closed under water to avoid contamination with surface films. The bottle was subsampled for a suite of analytes using a clean, 60 ml syringe triple rinsed with sample water. Dissolved inorganic and total dissolved nutrients were filtered through a 0.45 μm mixed cellulose ester (MCE) filter rinsed with 20 ml of sample water (discarded) before collection into triple rinsed 30 ml HDPE sample bottles. One subsample was assayed by flow injection analysis for dissolved inorganic nutrients using a Lachat Instruments QuikChem 8000 autoanalyzer for the analysis of NH_4, NO_3, NO_2, and SRP. Subsamples for TDN, TDP, TN and TP were assayed via two-step process: first water samples undergo a persulfate digest to convert all N into NO_3 and all P into orthophosphate; then the resulting digests were analyzed by automated colorimetry (Alpkem or Technicon) for nitrate-N and orthophosphate-P (Koroleff 1985).

Sediment Nutrients and Grain Size. Sediment characteristics and benthic chlorophyll *a* (benthic microalgae) were determined on sediment composite samples collected at the beginning and end of each macroalgae transect. Composites were comprised of ten sediment plugs (3 cm in diameter, 1 cm deep) collected at each end of the transect (20 total). The samples were collected downslope of the end of each transect in approximately 30 cm of water depth. Plugs were homogenized in sample bags and refrigerated until analysis. A subsample from each bag was collected in a 15 ml centrifuge tube and frozen for benthic chlorophyll *a* analysis. The remaining sediment was transferred to an aluminum dish, weighed wet and dried at 60°C to a constant weight and weighed dry. A subsample of dried sediment was ground with a mortar and pestle for analysis of percent total phosphorus (%TP), percent total nitrogen (%TN) and % organic carbon (%OC). Samples for %OC were acidified to remove carbonates; %OC and %TN were measured by high temperature combustion on a Control Equipment Corp CEC 440HA elemental analyzer at the Marine Science Institute, Santa Barbara. Sediment %TP were prepared using an acid persulfate digest to convert all P to orthophosphate that was then analyzed by automated colorimetry (Technicon) at the University of Georgia Analytical Chemistry Laboratory. The remainder of the sediment was reweighed dry, wet sieved through a 65 μm sieve, dried at 60°C to a constant weight, and weighed dry to determine grain size. Percent fines were calculated by difference from the total weight and the weight of the sieved portion. Benthic chlorophyll *a* samples were analyzed for chlorophyll *a* and phaeopigments as described above for phytoplankton.

Assessment Framework

We applied the thresholds from the proposed EU-WFD and ASSETS assessment frameworks without modification to macroalgae, phytoplankton, and dissolved oxygen data sets collected during the Bight'08 assessment of eutrophication. Relationships between other primary producer groups (microphytobenthos and *Ruppia spp.*) and nutrient enrichment have not been well characterized in the literature, so we opted to use only indicators for which assessment frameworks exist. However, a number of data integration decisions could be made in order to generate a final data set to which the thresholds could be applied (Table II-3; Figure II-2). We conducted sensitivity analyses on various approaches to data integration to understand how and to what extent it affected the ecological condition to which a segment was assigned. These approaches explored what data format to use, how data should be averaged to characterize the segment, and over what period of time should the data be integrated. An ecological condition category was assigned to each segment using each of the indicators, generating unique sets of ecological condition category assignments for each version of the data set. Options were evaluated and selected by the Bight'08 Eutrophication Assessment Workgroup for use in the final assessment of SCB estuaries. Single indicators scores were assigned to each estuary integrating over the entire year, as well as for individual sampling periods to represent percent of time the segment occupied each of the condition categories.

Macroalgal Abundance. Macroalgae are a natural component of shallow-water marine and estuarine soft-sediment communities; however excessive growth of opportunistic species may occur as a result of nutrient over-enrichment. Fong *et al*. (in Sutula 2011) provides a comprehensive review of macroalgae as an indicator of eutrophication and this information is briefly summarized here. An overabundance of macroalgae can have adverse effects on estuarine ecological health, including altered sediment and water column chemistry (e.g., hypoxia, ammonia, and sulfide toxicity), smothering of seagrass and reef habitats, and adverse effects to benthic invertebrates (Soulsby *et al.* 1982, Raffaelli *et al.* 1991, Cardoso *et al.* 2004a, Cardoso *et al.* 2004b). For these reasons, macroalgae has been identified as a relevant indicator to assess extent and magnitude of eutrophication in estuarine environments, particularly in California's bar-built lagoon and river mouth estuaries (Bricker *et al.* 2003, Scanlan *et al.* 2007, Sutula 2011).

Table II-3. Summary of data integration options that were evaluated through sensitivity analyses. Check mark and bold font designates those options that were used in final assessment of SCB Estuarine Eutrophication Assessment.

Issue	Indicator		
	Macroalgae	**Phytoplankton**	**Dissolved Oxygen**
Data format	• Wet weight ✓ **Dry weight**	✓ **Continuous** • discrete	✓ **Continuous**]
Data Integration Period	• Period with Highest Biomass/Cover ✓ **Average of two consecutive periods of highest biomass/cover** • Bight-wide Index period • Annual Average	• Maximum Instantaneous Value • Various percentiles (90th %percentile) ✓ **Annual Average**	• Minimum Instantaneous Value • 5th percentile ✓ **10th percentile** • 15th percentile
Smoothing applied to the data set	Transect values generated from: ✓ **Quadrat averages** • Quadrat percentile • Quadrat maximum	• Instantaneous ✓ **Daily running average**	• Instantaneous ✓ **Hourly running average**
Spatial extent over which data were integrated	Segment values generated from: ✓ **Average of Transects** • Maximum Transect • Percentile of Transect Data	✓ **Single Location**	✓ **Single Location**

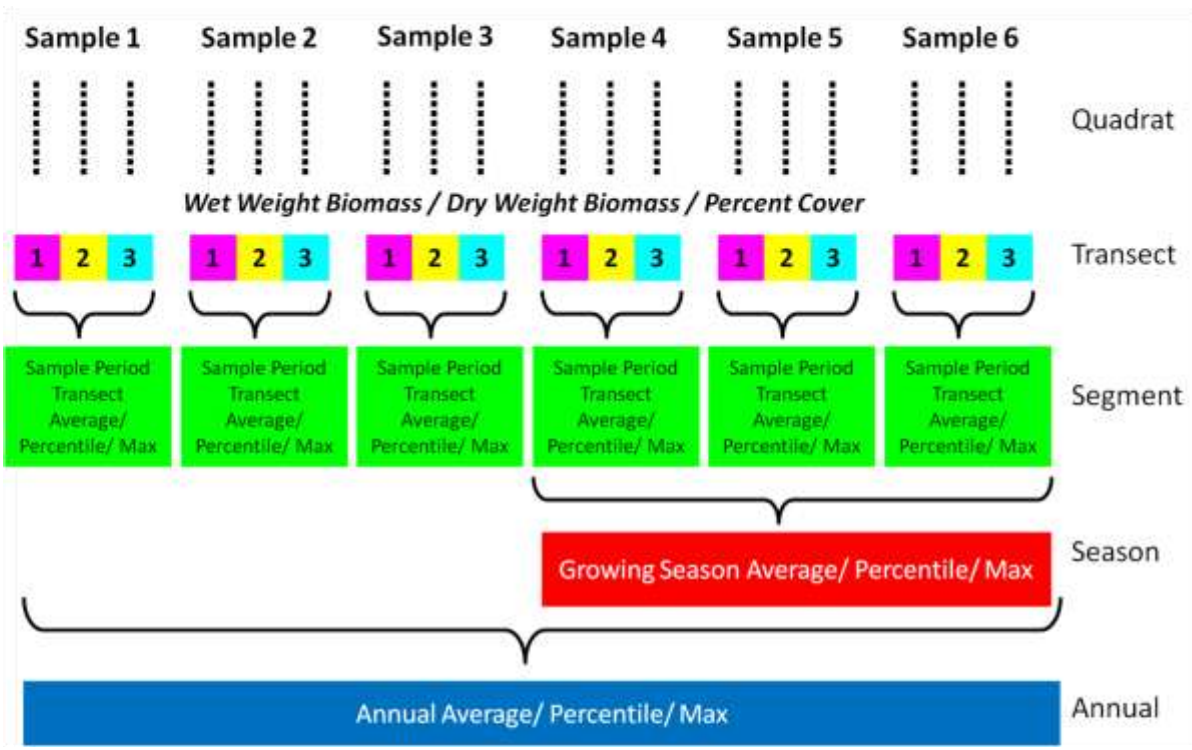


Figure II-2. Cartoon representing data integration options for macroalgae.

Studies supporting thresholds for macroalgal biomass and areal cover of macroalgae are limited (Bricker *et al.* 2003, Scanlan *et al.* 2007, Sutula 2011) and studies have only recently been conducted for California estuaries (Green 2011, Green *et al*. unpublished). The ASSETS framework, created in 2003, scores macroalgae as either a "problem" or "no problem" heuristically (Bricker *et al.* 2003), and thus was not considered further. The more recent Scanlan *et al* (Scanlan *et al.* 2007) framework proposed numeric thresholds used to categorize estuaries into five ecological condition categories based on a combination of biomass and cover. Biomass and cover thresholds were derived from a combination of published and unpublished studies as well as expert opinion. In order to use this framework to assess ecological condition, several choices for data management had to be addressed. Specifically, we investigated: 1) use of wet versus dry weight to assess biomass (data format), 2) spatial extent over which data were integrated, and 3) duration over which biomass and cover data were integrated (Table II-3).

Scanlan *et al*. (2007) state that thresholds for macroalgae biomass are based on wet weights for practical reasons because wet weights are a "much easier and less time-consuming determine than dry weights." However, wet weights have been found to be subject to error related to unequal amounts of water retained by individual samples and thus use of dry weight over wet weight has been supported by other investigators utilizing the proposed EU-WFD framework (Patricio *et al.* 2007). Therefore, we chose to convert Scanlan *et al*. (2007) thresholds for wet weight biomass to dry weight. In order to do this, we reasoned that the data supporting the development of the EU-WFD wet weight thresholds was based on large blooms of opportunistic macroalgae dominated by species of sheet and tube forming *Ulva.* Although there was no significant difference between the wet: dry weight ratios of red vs. green algae, we utilized the median dry: wet weight ratio of all ulvoid biomass samples to convert the Scanlan *et al*. (2007) wet weight thresholds to dry weight. The framework to score ecological condition using macroalgae biomass and cover using is presented in Table II-4, giving both wet (original) and dry weight (converted) biomass thresholds. These dry weight thresholds were applied to the Bight'08 data set for scoring overall condition, and the comparison to assessments made with wet weights is discussed.

Table II-4. Categorization of estuarine ecological condition with respect to eutrophication based on dry weight biomass and percent cover values used for the Bight Estuarine Eutrophication Assessment.

Biomass (g ww m^{-2})	Biomass (g dw m^{-2}) *(converted)*	Percent Cover				
		≤5%	> 5%	> 15%	> 25%	> 75%
> 3000	> 415	Moderate	Low	Very Low	Very Low	Very Low
1000 - 3000	140 - 415	Moderate	Moderate	Low	Very Low	Very Low
500- 1000	70 - 140	High	Moderate	Moderate	Low	Low
100 - 500	15 - 70	Very High	High	High	Moderate	Low
< 100	≤ 15	Very High	High	High	Moderate	Moderate

*"Very High" = nearly undisturbed conditions; "High" = slight change in composition and/or biomass; "Moderate"= moderate change in composition and/or biomass; "Low" = major change in biological communities; "Very Low" = severe change in biological communities.
** Moderate or worse status is considered a problem area for the WFD.

The EU-WFD framework does not specify how data should be collected, although Scanlan *et al*. (2007) proposed two survey methods for assessing macroalgae blooms (transects versus aerial estimates) and both options generated consistent results (Patricio *et al.* 2007). Our data was collected in discrete quadrats within three transects per segment. Quadrat data was averaged into transect values as this is commonly accepted as a good representation of spatially patchy algal biomass and cover (Kennison *et al.* 2003). We also considered how to integrate transect data into a segment value, including 1) average of the three transects and 2) use transect with highest biomass/cover. We conducted sensitivity analysis on both options, but chose to use the average of the three transects to characterize the segment per the decision of the Estuarine Eutrophication Workgroup.

We also considered how duration incorporated into the assessment. The EU-WFD indicates that sampling should be conducted at the peak of biomass and recommends that multiple samplings occur to ensure that the "peak" is captured, but does not have recommendations for how duration should be quantified. We considered: 1) peak biomass and cover, 2) annual average, and 3) average of two consecutive periods of highest biomass and cover. Estuaries in southern California have peak macroalgea biomass and cover at different times throughout the year (Section III). Furthermore, recent research has shown that biomass and cover values similar to what we observed are detrimental to benthic infauna after eight weeks of exposure (Green 2011). We conducted sensitivity analysis on all three options, but chose to apply thresholds to the average values of the two consecutive highest periods of biomass and cover (representing at least 8 weeks of coverage).

Phytoplankton Biomass. Water column chlorophyll *a* is a common indicator of phytoplankton biomass in inland, estuarine and coastal waters (Cloern 2001, Bricker *et al.* 2003). Elevated chlorophyll *a* concentrations reduce light to benthic primary producers, including seagrass and microphytobenthos, create conditions leading to hypoxia, and are associated with the promotion of algal blooms of undesirable or harmful algal blooms (see (Sutula 2011) for comprehensive review). The EU WFD is considering phytoplankton assessment frameworks available from ASSETS (Bricker *et al.* 2003) and from the French Research Institute for Exploitation of the Sea (IFREMER) for use in French Mediterranean Lagoons (Souchu *et al.* 2000, Zaldivar *et al.* 2008). Both frameworks conceptually use shading of seagrass habitats as the basis for threshold selection. However, the averaging period of the thresholds is slightly different. ASSETS aimed to address the highest concentrations by using the 90th percentile of annual chlorophyll *a* concentration data and an estimate of the frequency of occurrence (Bricker *et al.* 2003). The IFREMER framework addresses captures the presence of chronic blooms by using the annual average of chlorophyll *a*, where data is collected at a minimum of once per month (Souchu *et al.* 2000, Zaldivar *et al.* 2008) and for this reason uses lower thresholds than ASSETS. We compared the results from the two approaches (Table II-5), but chose to apply the IFREMER framework to make our assessment of SCB estuaries as thresholds from French Mediterranean lagoons are likely to be more applicable to southern California estuaries.

Table II-5. Thresholds for phytoplankton chlorophyll *a* used by Souchu *et al*. (2000) for the assessment ecological condition of French Mediterranean lagoons (as given in Zalidvar *et al*. 2008), and thresholds and ranges of chlorophyll *a* as defined by ASSETS (Bricker *et al*. 2003).

EU-WFD: IFREMER		ASSETS	
Mean Annual Chl *a* Concentration	**Ecological Condition Category**	**90th percentile of Annual Chl *a* Concentration**	**Range Definition***
0 - 5 µg Chl *a* L^{-1}	Very High	0 – 5 µg Chl *a* L^{-1}	Low Levels
5 - 7 µg Chl *a* L^{-1}	High	-	-
7 - 10 µg Chl *a* L^{-1}	Moderate	5 - 20 µg Chl *a* L^{-1}	Medium Levels
10 - 30 µg Chl *a* L^{-1}	Low	20 - 60 µg Chl *a* L^{-1}	High Levels
>30 µg Chl *a* L^{-1}	Very Low	>60 µg Chl *a* L^{-1}	Hypereutrophic

*We imposed the EU-WFD color scheme on ASSETS chlorophyll *a* ranges to compare results between the two frameworks.

To investigate sensitivity of temporal integration period on the results generated using both the IFRMER and ASSETS thresholds, we compare ecological condition categories assigned using: 1) annual average and 2) 75th percentile and 90th percentile of peak bloom period. In addition, we collected water column chlorophyll *a* grab samples with the intent of calibrating continuous chlorophyll *a* fluorescence data collected via data sondes, so the calibrated continuous data were used for this purpose. However, we explored how the use of continuous versus grab sample data effected the categorization of estuaries. We also compared the results of smoothing the continuous data with a running daily average (to eliminate high frequency noise) with the use the instantaneous (15-minute) data.

While we do not have spatial data with which to enhance our assessment of ecological condition using chlorophyll *a* data, both ASSETS and the IFREMER frameworks do recognize that chlorophyll *a* concentrations must be significant not only in terms of duration but also in terms of spatial extent (Bricker *et al.* 2003, Zaldivar *et al.* 2008). ASSETS includes an assignment of a spatial coverage category (high, moderate, low, or very low) in addition to magnitude and frequency categories. Neither framework outlines how much spatial coverage is required to make an accurate assessment.

Duration of phytoplankton blooms is only indirectly addressed in both the IFREMER and ASSETS assessment frameworks through the use of annual averages and percentiles of annual data respectively. We investigated the how duration could be more directly addressed through application of the IFREMER thresholds to each daily average of chlorophyll *a.* We calculated the consecutive number of days each estuary was in a category of moderate or worse condition as a measured of bloom duration.

Dissolved Oxygen Concentration. Low dissolved oxygen (DO) has direct effects on the reproduction, growth and survival of pelagic and benthic fish and invertebrates (USEPA 2000, Bricker *et al.* 2003, Best *et al.* 2007). Thresholds for assessment of effects of DO are derived from criteria deemed to be protective of the most sensitive species from acute and chronic exposures to low dissolved oxygen. Results from the Bight'08 estuarine eutrophication assessment indicate that bottom water dissolved oxygen concentrations were strongly correlated with sediment organic carbon and total nitrogen (Section IV). This relationship suggests that dissolved oxygen is an indicator of long-term nutrient and organic matter loading into an estuary.

Both ASSETS and the proposed framework for the EU-WFD include dissolved oxygen as an indicator of eutrophication (Bricker *et al.* 2003, Best *et al.* 2007). Thresholds for both frameworks are based on observed impacts of hypoxia on benthic and demersal fauna as well as expert opinion and are targeted to be relevant in a wide range of estuarine environments (Table II-6). These thresholds are similar to those proposed by an expert panel assembled to determine relevant thresholds for dissolved oxygen on California species, including salmonids found in some southern California estuaries (Sutula *et al*. 2012). In addition, the proposed framework for the EU-WFD (Best *et al.* 2007) has the advantage of incorporating the salinity effects on oxygen solubility. We present a comparison of ASSETS versus Best *et al*. (2007), but chose to apply the EU-WFD framework to our dissolved oxygen datasets to generate an assessment for SCB segments.

Table II-6. Thresholds for dissolved oxygen concentration proposed for the EU-WFD (Best *et al*. 2007) and ASSETS (Bricker *et al*. 2003) assessment frameworks.

Framework	Proposed Ecological Status	Freshwater	Marine	Rational
EU-WFD	Very High	≥ 7 mg L^{-1}	≥ 5.7 mg L^{-1}	Supports all life stages of salmonids and transitional fish
	High	5 - 7 mg L^{-1}	4.0 - 5.7 mg L^{-1}	Supports presence of salmonids and transitional fish
	Moderate	3 - 5 mg L^{-1}	2.4 - 4.0 mg L^{-1}	Supports most life stages of non-salmonid adults
	Low	2 - 3 mg L^{-1}	1.6 - 2.4 mg L^{-1}	Supports presence of non-salmonids, poor survival of salmonids
	Very Low	< 2 mg L^{-1}	< 1.6 mg L^{-1}	No salmonids, marginal survival of resident species
ASSETS*	High	> 5 mg L^{-1}		Protective of fish and invertebrate species
	Biologically Stressful	2 - 5 mg L^{-1}		Field and laboratory observations show stress responses in invertebrate and fish fauna
	Hypoxia	0 - 2 mg L^{-1}		Significantly reduced benthic macroinfauna, epifauna and demersal species

*We imposed the EU-WFD color scheme on ASSETS dissolved oxygen ranges to compare results between the two frameworks.

Both ASSETS and EU-WFD (Bricker *et al.* 2003, Best *et al.* 2007) utilize a percentile approach to data integration, calculated by ranking the continuous data from lowest to highest value, and applying the percentile. The EU-WFD applies a fifth percentile and ASSETS a tenth percentile; thus the 5th percentile of 9 months of continuous DO data equates to approximately 2 weeks below a designated threshold, although this time is not necessarily consecutive and could indicate multiple events or a single long event. The use of the percentile approach is a step towards integration of duration and frequency of low DO events, although the length of events and the frequency are not accounted for in this approach. However, ASSETS includes a frequency category in addition to categories for magnitude and spatial extent and the EU-WFD recommend using Fundamental Intermittent Standards (FIS) and a return period in more sensitive habitat that sets a minimum threshold for dissolved oxygen, a duration, and a return period. The EU-WFD proposed a second tier FIS for "high" ecological condition that dissolved oxygen should not fall below 2 mg L^{-1}, more frequently that once every 6 years over a 6 hour tidal cycle and for "moderate" status not more than once every 3 years (Best *et al.* 2007). Because we only have one year of data, we could not include this second tier standard.

We collected data from a single site, in bottom water, in a segment of the estuary that was most likely to experience symptoms of eutrophication due to higher residence times. Thus, our dissolved oxygen data is likely to represent the lowest levels of dissolved oxygen for each of the estuaries. For this reason we opted to use the EU-WFD thresholds but to apply them to the 10th percentile of the data (thresholds should be exceeded 90% of the time), rather than the 5th percentile. To investigate the effect of using different percentiles on the resulting ecological condition category, we applied the thresholds for both the EU-WFD and ASSETS to the 5th, 10th, and 15th percentile of data for each segment. In terms of data format, we chose to apply an hourly running average filter to the data rather than use the instantaneous data to eliminate high frequency noise for the assessment. We compared these results to those generated with instantaneous data to determine the effect of the filter on the resulting ecological condition category.

While we do not have spatial data with which to enhance our assessment of ecological condition using dissolved oxygen concentrations, both ASSETS and the IFREMER frameworks do recognize that hypoxic events must be significant not only in terms of duration but also in terms of spatial extent (Bricker *et al.* 2003, Best *et al.* 2007). ASSETS includes an assignment of a spatial coverage category (high, moderate, low, or very low) in addition to magnitude and frequency categories. However, neither framework outlines how much spatial coverage is required to make an accurate assessment. The EU-WFD dissolved oxygen framework proposes two types of data collection, spot measurements made frequently at many sites throughout a water body as well as continuous monitoring from a limited number of strategically placed data sondes. The proposed thresholds have been applied to both types of data, although there are occasionally differences in condition category using the different data in the same system (Best *et al.* 2007, Greenwood *et al.* 2010). Each data type has its potential pitfalls: spot measurements may miss periods of high stress and continuous monitors of a small number of sites may not reflect the general condition of the estuary (Best *et al.* 2007).

Duration of low oxygen events is captured as an integrated measure over time in both the EU-WFD and ASSETS assessment frameworks through the use of percentiles. We calculated the consecutive number of days each segment was in a category of moderate or worse DO condition as a measure of low DO event duration. We also applied the WFD thresholds to the hourly averaged dataset for all segment sites to determine how the condition of the estuary varies on short timescales throughout the year**.**

Multiple Lines of Evidence. ASSETS and the EU-WFD have different approaches to dealing with multiple lines of evidence. ASSETS is truly a multi-metric approach where indicators of response are fully integrated with metrics of pressure and state to generate a complete picture of ecological condition in the present as well as projected into the future (Bricker *et al.* 2003). The EU-WFD is typically applied as a "one out, all out", where the lowest score for any single element determines the ecological condition category for the estuary overall (Borja *et al.* 2006, Zaldivar *et al.* 2008). Though recently calls have been made to integrate elements of the EU-WFD into a more unified, multi-metric approach to categorizing ecological condition (Borja *et al.* 2009a, Borja and Rodriguez 2010, Borja *et al.* 2011). We compare the designated categories for each segment to see how well they agreed and discuss how multiple lines of evidence might be utilized for SCB estuaries.

Results

Extent and Magnitude of Eutrophication in Southern California Bight Estuaries

In this section we describe the results for each indicator individually for each estuarine segment and then as multiple lines of evidence for eutrophication, comparing the degree to which the indicators agree.

Macroalgal Abundance. According to the EU-WFD, 78% of segments were categorized as having moderate or worse ecological condition (Figure II-3A). However, a substantial fraction of estuaries fell in the "moderate" category (37%) and placement in the "moderate" ecological condition category was largely driven by cover between 25% and 50% and biomass less than 70 g dw m^{-2} (Figure II-3B). Peak season average biomass and cover ranged from 0 g dw m^{-2} and 0 % cover to 295 g dw m^{-2} and 65 % cover (site with highest biomass) and 91 g dw m^{-2} and 93% cover (site with highest cover).

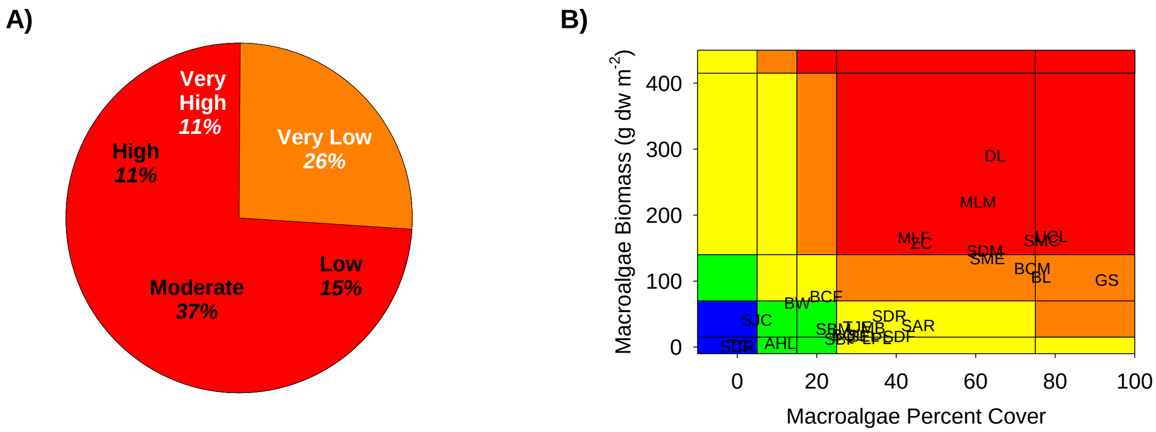


Figure II-3. Percent of segments falling into each ecological condition category based on WFD integrated score of macroalgae biomass and cover (A), and Categorization of SCB segments according to ecological condition based on distribution of macroalgal biomass and cover (B).

The percentage of time any given system spent in each ecological condition category was highly variable from site to site (Figure II-4). Some segments had chronically high biomass and cover scoring moderate or worse for > 80% of the year (e.g., DL, GS, UCL, MLM, BL), whereas other sites had more episodic blooms lasting only one sampling period (e.g., ZC, SJC, SBF). For most segments, the overall score is driven by more than one period of moderate or worse ecological condition; however for two segments, the score is largely driven by high biomass and cover in only a single sampling period (ZC, SBF).

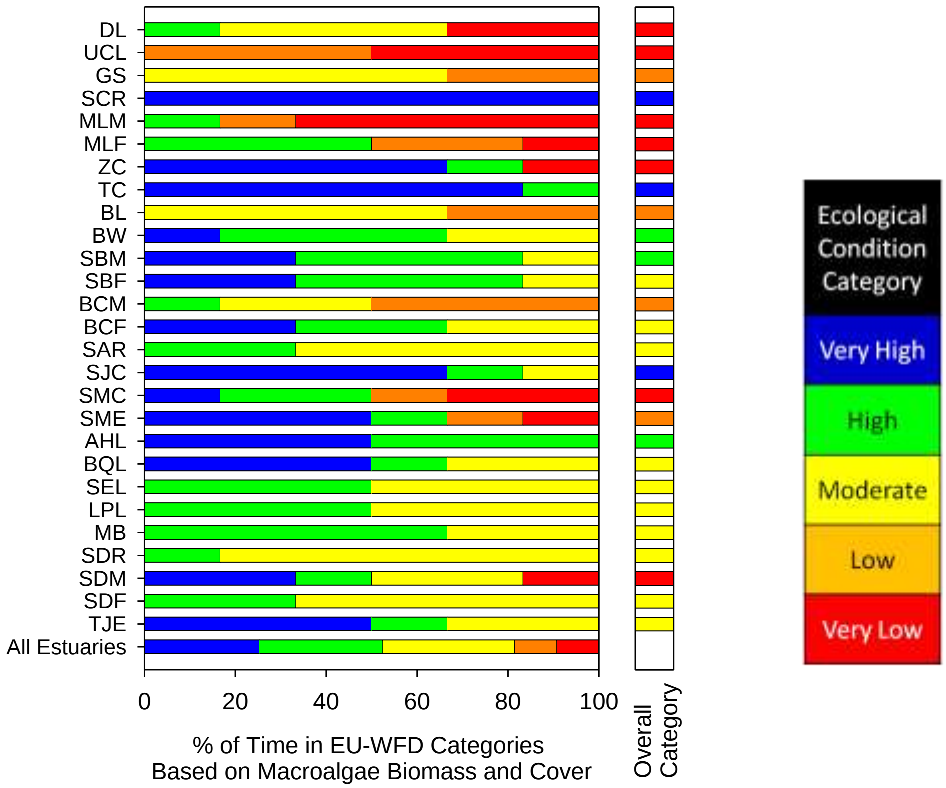


Figure II-4. Percent of total time each segment spends in any given ecological condition category and overall peak season score.

Because we sampled every other month, we cannot assess bloom duration for a period of less than 8 weeks; however, fifteen of the segment sites (55%) had macroalgae biomass and cover that placed them in a category of moderate or worse ecological condition for two or more consecutive periods (> 8 weeks). However, for most of these estuaries, the consecutive periods are in the "moderate" category, rather than low or very low categories. Thirty percent of segments had two or more periods of low/very low ecological condition and 11% of the segments had two or more consecutive periods of very low ecological condition as defined by macroalgae biomass and cover (Figure II-5). For the SCB segments sampled, 37% of sites had moderate or worse biomass for 12 or more weeks (3 or more consecutive periods) and 26% had moderate or worse biomass for longer than 20 weeks (5 or more consecutive periods). Three sites (11% of segments) had continuous coverage of moderate or worse ecological condition throughout the sample year and one of these sites was continuously in a low or very low condition according to the WFD framework.

Phytoplankton Biomass. It should be noted that phytoplankton as an indicator of eutrophication is only applicable in systems with significant subtidal area, and 26% of SCB estuaries have more intertidal area than subtidal area (Table II-1; 7 segments: BW, LPL, MLF, MLM, SEL, SJC, TJE, 26%). Therefore, although it was applied to all segments, phytoplankton may not be a relevant indicator in these systems.

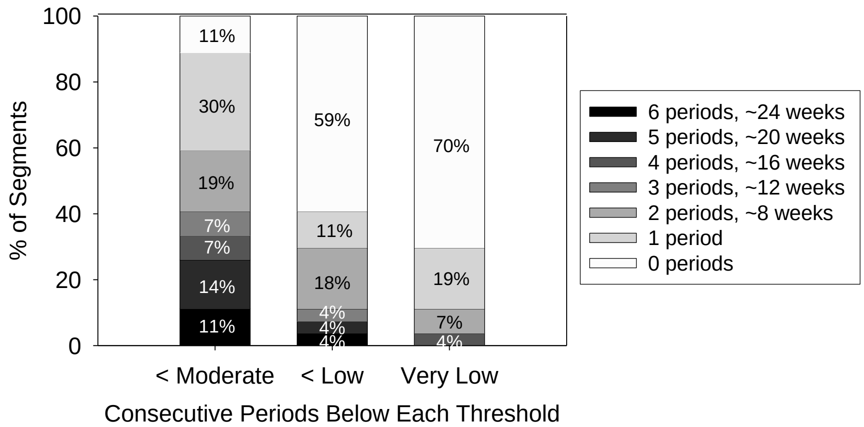


Figure II-5. Duration of macroalgae bloom events resulting in a category of moderate or worse (left bar), low or worse (middle bar), and very low (right bar) ecological condition in each estuary.

Annual average chlorophyll *a* concentrations ranged from 0.5 to 42 µg L^{-1}. Of the three biological response indicators, phytoplankton biomass has the fewest number of segments falling in an IFREMER category of moderate or worse (39%; Figure II-6A). Eleven percent of segments were scored in the "moderate" category. One segment had a very high annual average of chlorophyll *a* (Santa Clara River Estuary, SCR; Figure II-7). This system and San Juan Creek (SJC) had little macroalgae (categorized as "very high ecological condition" based on macroalgae biomass and cover), but high chlorophyll *a* (categorized as "very low" for SCR and "low" for SJC).

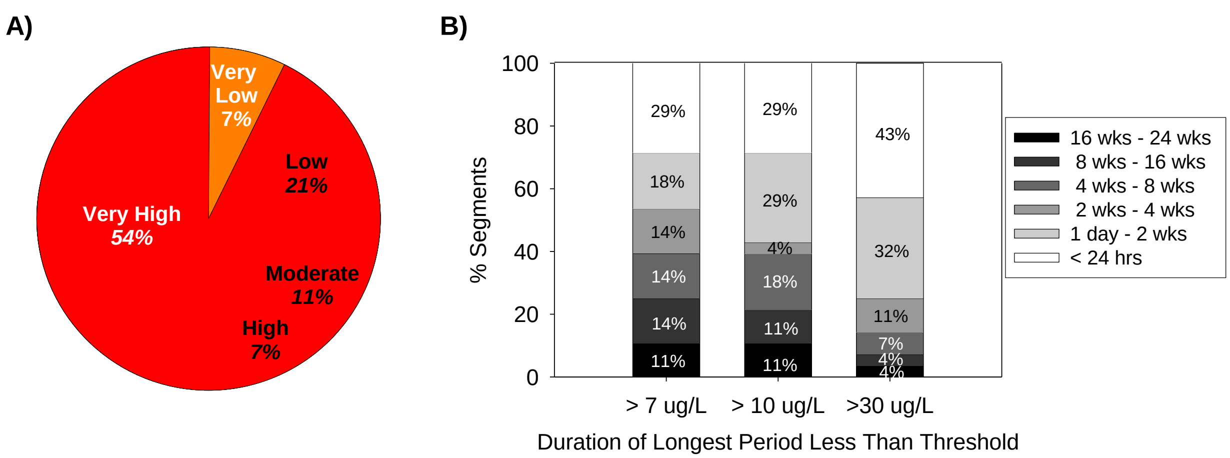


Figure II-6. Percent of segments falling into each ecological condition category based on chlorophyll *a* (A). Duration of phytoplankton bloom events resulting in a category of moderate or worse (left bar), low or worse (middle bar), and very low (right bar) ecological condition in each estuary (B).

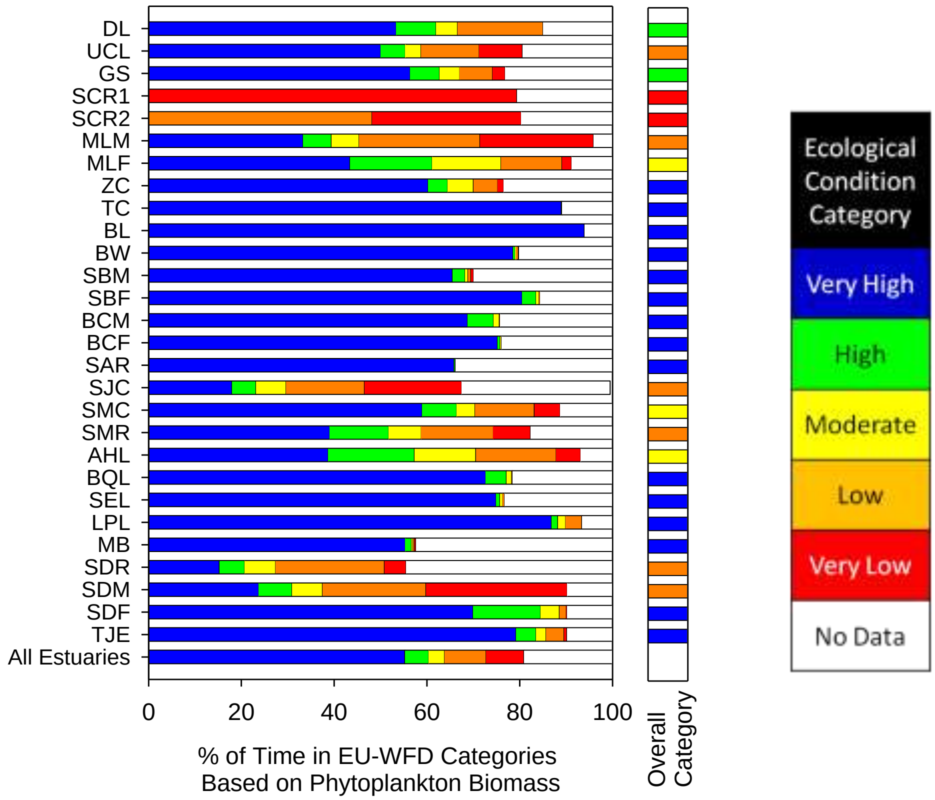


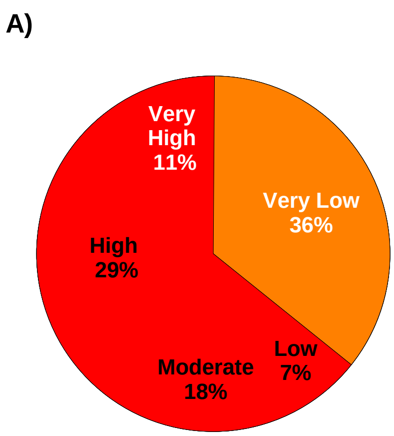
Figure II-7. Percent of time each segment spends in any given ecological condition category and overall annual average score (time is not necessarily consecutive).

Based on an annual average, the percentage of time any given system spent in each ecological condition category was variable, though one third of segments spent less than 10% of the time in an ecological condition category of moderate or worse (Figure II-7). Some segments had chronically high chlorophyll *a* scoring moderate or worse for over half of the year (e.g., SCR, SDM, SJC, SDR, MLM), whereas other sites have more episodic blooms lasting only a few weeks (e.g., BW, SBM, SBF,BCM, BCF, SEL, MB). All segments scoring an overall category of moderate or worse had daily average chlorophyll *a* concentrations in the moderate or worse category for >25% of the year.

With respect to bloom duration, 40% of segments had continuous phytoplankton blooms greater than 7 and 10 $\mu g\ L^{-1}$ for longer than 1 month (Figure II-6B). Twenty-five percent of segments had biomass greater than 7 $\mu g\ L^{-1}$, and 22% greater than 10 $\mu g\ L^{-1}$ for longer than 2 months. For biomass greater than 30 $\mu g\ L^{-1}$, 15% of segments exceeded this threshold continuously for 1 month and 8% for 2 months. Some systems that fell in a "high" or "very high" ecological condition category using an annual average of chlorophyll *a,* had concentrations above 7 $\mu g\ L^{-1}$ for periods of short duration. These short-term events were typically less than 1 month in duration.

Data gaps were a concern for some of the segments. Five segments had data sets that were less than 80% complete due to logistical issues and probe failures (MB, SDR, SAR, SJC, SBM). However, data gaps for all but two of the systems were spread evenly throughout the year and thus do not likely impact the score. For two of the sites (MB and SDR), sondes were not deployed until April so the data gap is largely at the beginning of the data set. Most segments did not experience a bloom until late spring/early summer and the data gap for these two sites could potentially result in score that would place them in a lower ecological condition category than would have been achieved had the sondes been deployed for the full deployment period because the measured, higher chlorophyll concentrations of the summer would not be averaged with unmeasured, lower chlorophyll values at the beginning of the year when the "annual" mean is calculated.

Dissolved Oxygen. For dissolved oxygen, 10th percentile concentrations over the 9-month period of January - October 2009 ranged from 0 mg L^{-1} to 7 mg L^{-1}. Sixty-one percent of segments fell into an ecological condition category of moderate or worse using dissolved oxygen concentration as an indicator (Figure II-8A). Of these systems, the greatest percentage fell into a category of "very low" ecological condition (36%). Thresholds for dissolved oxygen are based on observed impacts of hypoxia on resident species (Best *et al.* 2007). Forty percent of segments fell in a high or very high condition class which protects adult salmonid survival, and 11% of segments fell in the very high condition category which protects all life stages of salmonids. However, not all segments would be expected to support salmonid populations.



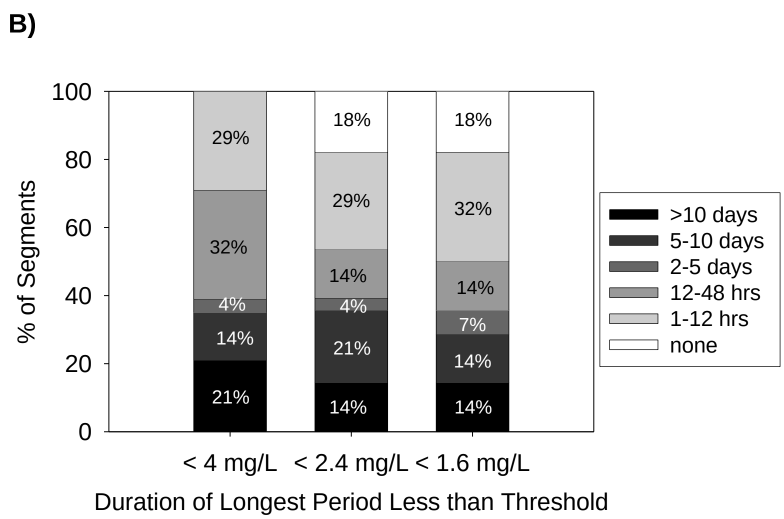


Figure II-8. A) Percent of segments falling into each ecological condition category based on dissolved oxygen concentrations, B) Duration of hypoxia resulting in a category of moderate or worse (left bar), low or worse (middle bar), and very low (right bar) ecological condition in each estuary.

The percentage of time any given segment spent in each ecological condition category was variable from site to site, and every site spends some time in a category of moderate or worse (Figure II-9). Most segments fall into the moderate or worse category for only a portion of the diel cycle (day-night) and the percentage of time in a moderate or worse ecological condition category is reflective of many consecutive nights of low dissolved oxygen concentration rather than a single continuous time period of low dissolved oxygen. However, for some segments continuous low dissolved oxygen events exceeding diel cycles was a problem (e.g., SCR, DL, GS, UCL).

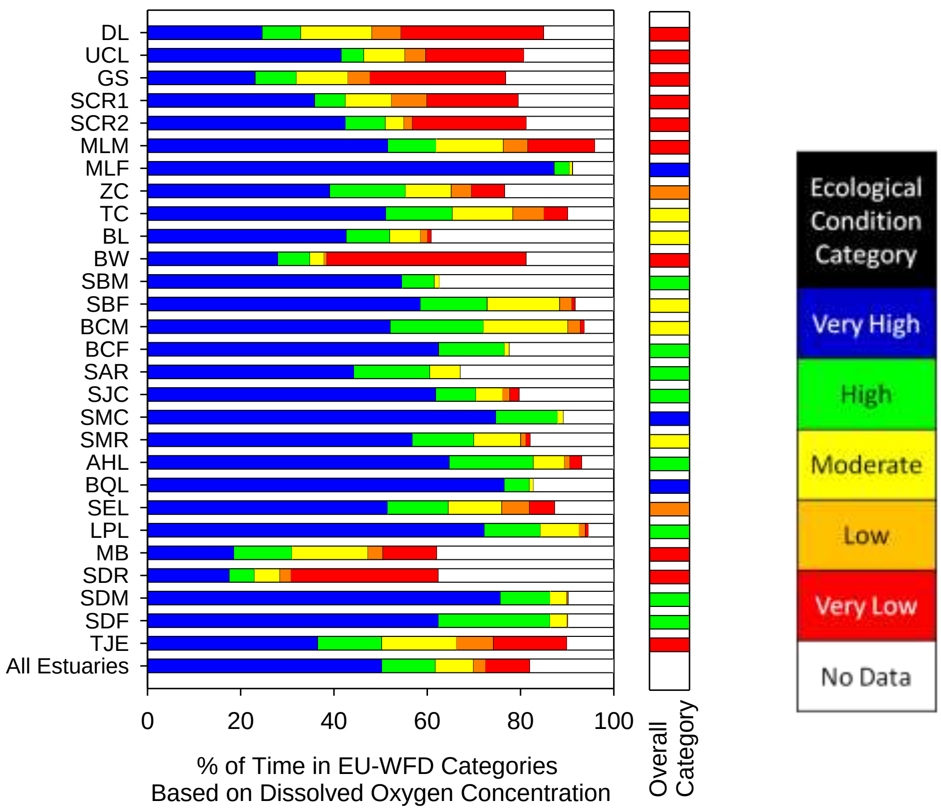
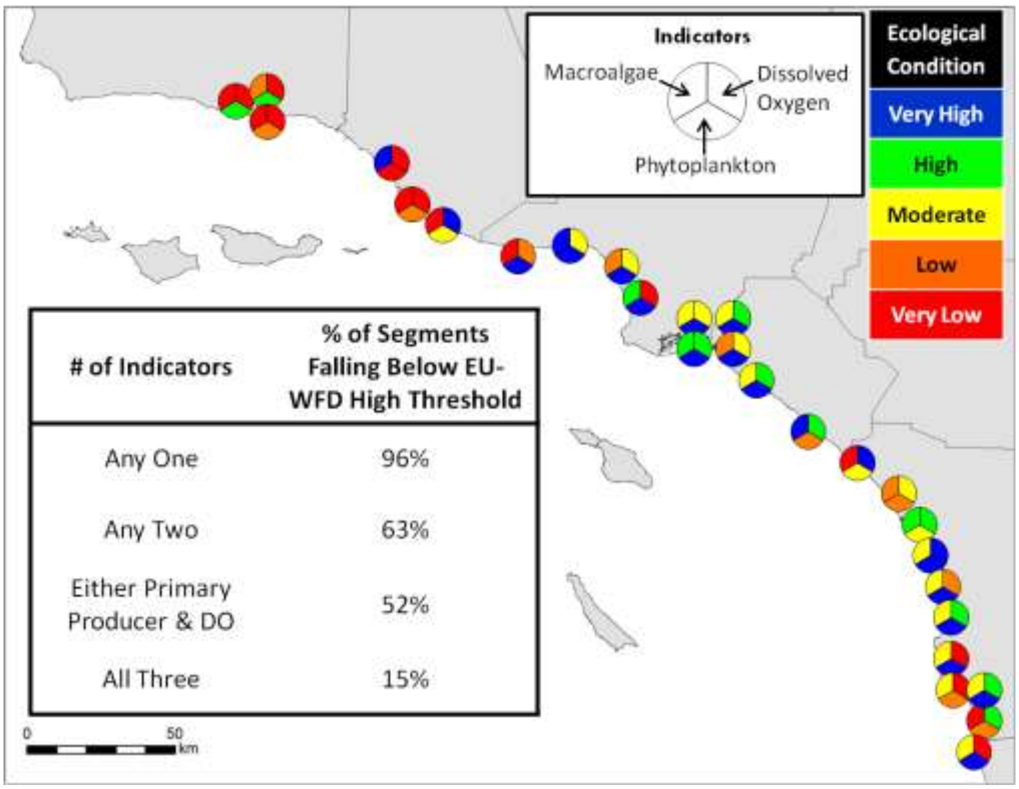


Figure II-9. Percent of time each segment spends in any given ecological condition category and overall 10th percentile score (time is not necessarily consecutive).

All SCB segments had some period of time less than the moderate threshold of 4 mg L^{-1}, and 82% of sites spend some time below the low and very low thresholds (2.4 mg L^{-1} and 1.6 mg L^{-1} respectively). Though for 29% of segments, the longest continuous period less than the moderate and low thresholds was 12 hours or less (Figure II-8). For longer duration events, 35% of segments had concentrations less than 4 and 2.4 mg O_2 L^{-1} for longer than 5 days. Twenty-eight percent of SCB segments had concentrations continuously less than 1.6 mg O_2 L^{-1} for longer than 5 days and 14 % for longer than 10 days.

As with phytoplankton, data gaps in the continuous dissolved oxygen dataset were a concern for some of the segments, particularly MB and SDR due to the delay in deployment described above. For most segments hypoxia is not a problem until late spring/early summer and the data gap for these two sites could potentially result in score that would place them in a lower ecological condition category than would have been achieved had the sondes been deployed for the full deployment period because the higher dissolved oxygen concentrations in the winter would not be included in the ranked data.

Multiple Lines of Evidence. The three indicators of biological response to eutrophication did not necessarily agree on the ecological condition of any given estuarine segment (Figure II-10). All but one segment (96% of segments) were assigned an ecological condition class of moderate or worse based on any one indicator, a condition category that would require management action according to the WFD framework (Zaldivar *et al.* 2008). This percentage drops to 63% if any two indicators are considered and to 53% if the two indicators must include one of the primary producers and dissolved oxygen concentration. Fifteen percent of segments (MLM, SDR, SMR, UCL) fell in a category of moderate or worse in all three biological response indicators.



# of Indicators	% of Segments Falling Below EU-WFD High Threshold
Any One	96%
Any Two	63%
Either Primary Producer & DO	52%
All Three	15%

Figure II-10. Ecological condition category determined for each estuarine segment using three response indicators of eutrophication: macroalgae, phytoplankton and dissolved oxygen concentration. Summary table shows the percent of segments falling below the "high" threshold for one, two or three indicators.

Segments in a Regional Context. Eutrophication in any given segment can also be assessed relative to other estuaries in the region by ranking segments based on the magnitude of response of the indicators. To this end, we ranked segments from best ecological condition (lowest macroalgae and phytoplankton biomass, highest dissolved oxygen) to worst ecological condition (highest macroalgae and phytoplankton biomass, lowest dissolved oxygen) for each indicator. We generated an overall rank from best (#1) to worst (#27) from the average of the rank for the dominant primary producer (primary symptom) and dissolved oxygen (secondary symptom; Table II-7). In addition to putting the segments into a regional context, this exercise helps to highlight which indicators are relevant in specific systems and how site specific factors can mediate the response of some indicators. For example SCR and SJC ranked among the segments with highest ecological condition based on macroalgae, but among the lowest for phytoplankton and low overall. Similarly, a large number of systems ranked among the lowest for macroalgae and highest for phytoplankton (e.g., SME, SDM, SCM, ZC, SBF). However, rankings based on phytoplankton are somewhat arbitrary because estuaries ranked 1 through 15 are all in a "very high" ecological condition category according to the EU-WFD. There are also two sites with that rank among the lowest ecological condition for both primary producers but among the highest for dissolved oxygen (MLF, SMC).

Sensitivity of Results to Threshold, Data Format and Spatio-temporal Integration

A number of segments were on the borderline of thresholds that would place them in a different category, which would make them prone to reclassification given a different data management regime or new threshold. This would be particularly important for segments falling in the "moderate" category, since the threshold between "high" and "moderate" drives management action according to the EU-WFD. To investigate the sensitivity of ecological condition category to changes in data format, data integration, and threshold selection, we compared the results from the Bight'08 assessment as described above to results generated using a different data management options and to results generated using thresholds from the ASSETS assessment framework. Results are summarized in Table II-8, with detail given in Tables II-9, II-10 and II-11.

For macroalgae, the outcome of the assessment was sensitive to data format, spatial and temporal integration of the data, though specific segments were more sensitive than others (Table II-9). Use of wet weights versus dry weight had a significant effect on individual segments, with 19% segments increasing an ecological condition score and 11% decreasing. Using the transect with the highest biomass and cover decreased the ecological condition score of 44% of segments and for 11% of these segments the score changed by 2 or more categories. Similarly, if an annual average of all segment data is used, 41% segments increase in ecological condition score and one segment decreases, whereas if the single period of highest biomass and coverage (maximum period) is used 7% segments increase in score and 22% decrease in score. Use of an annual average of segment values (annual average) generated from an average of the three transects (transect average) results in the maximum number of segments being in the highest possible ecological condition category, although 56% segments will have no change in ecological condition category. Use of the single period of highest biomass and cover (maximum period) generated from the transect with highest biomass and cover (maximum transect) results the lowest possible score for each segment, although 37% segments will have no change in condition category.

Table II-7. Ranks of segments. Each indicator is ranked from 1 (best) to 27 (lowest ecological condition); peak season macroalgal abundance, and phytoplankton (as an annual average) is ranked from lowest to highest abundance, dissolved oxygen is ranked from highest to lowest value at the 10th %tile. Overall rank is determined from the average of the dissolved oxygen and the lowest ranked of either algal group. The colors represent the EU-WFD score assigned to the segment for each indicator: very high (blue), high (green), moderate (yellow), low (orange), very low (red).

Overall rank		Segment	Segment Rank by Indicator*		
			Peak Season Macroalgal Abundance	Annual Mean Phytoplankton Biomass	10th Percentile Dissolved Oxygen
Highest Condition ---------------------------------------> Lowest Condition	1	Batiquitos Lagoon	7	10	3
	2	Seal Beach	5	12	4
	3	Los Penasquitos Lagoon	6	7	10
	4	Bolsa Chica	15	11	5
	5	Mugu Lagoon	18	19	1
	6	San Diego Bay	10	15	7
	7	Santa Ana R. Wetlands-Diked	14	2	9
	8	Seal Beach- Diked	9	8	14
	9	San Elijo Lagoon	8	5	17
	10	San Mateo Lagoon	24	18	2
	11	Topanga Lagoon	2	13	16
	12	Tijuana River Estuary	11	1	19
	13	San Diego Bay- Diked	22	24	6
	14	Agua Hedionda Lagoon	3	20	11
	15	Ballona Lagoon- Diked	19	3	13
	16	Santa Margarita Estuary	20	21	12
	17	San Juan Creek	4	25	8
	18	Zuma Lagoon	17	14	18
	19	Bolsa Chica- Diked	21	4	15
	20	Ballona Wetlands	13	6	26
	21	Mission Bay	12	9	27
	22	Goleta Slough	23	17	23
	23	Mugu Lagoon - Diked	26	26	20
	24	San Diego River	16	23	24
	25	UCSB Campus Lagoon	25	22	22
	26	Santa Clara River	1	27	21
	27	Devereux Lagoon	27	16	25

*Not all indicators are relevant in all segments, relative ranks for indicators may not reflect better ecological condition if the indicator is not relevant in a particular system.

Table II-8. Number of segments that change ecological condition class due to a change in data format, framework, or integration relative to that used to classify extent and magnitude of SCB estuarine segments in the Bight'08 regional assessment. PS = Peak Season. MP = Max period.

Indicator and Comparator	Data Format	Framework	Data Management	# Segments that change condition category				
				no change	+1	+2 or more	-1	-2 or more
Macroalgae WFD Framework Dry Biomass Peak Season Average/ Transect Average	Dry Biomass	EW-WFD	Annual Avg/Transect Avg	15	9	2	1	0
		EW-WFD	PS Avg/Transect Max	15	0	0	9	3
		EW-WFD	MP/ Transect Avg	22	0	0	4	1
		EW-WFD	MP/ Max Transect	10	0	0	13	4
	Wet Biomass	EW-WFD	Annual Avg/Transect Avg	14	8	4	1	0
		EW-WFD	PS Avg/ Transect Avg	19	5	0	3	0
		EW-WFD	PS Avg/ Transect Max	12	2	0	10	3
		EW-WFD	MP/ Transect Avg	19	2	0	6	0
		EW-WFD	MP/ Max Transect	10	0	0	12	5
Phytoplankton IFREMER Framework Annual Average of Daily Averages	Daily Averages	EW-WFD	75th %tile	22	1	0	5	0
		EW-WFD	90th%tile	16	0	0	9	3
		ASSETS	Annual Avg	21	5	0	2	0
		ASSETS	75th %tile	20	5	0	2	1
		ASSETS	90th%tile	18	2	0	5	3
	Instant-aneous	EW-WFD	Annual Avg	27	0	0	1	0
		EW-WFD	75th %tile	23	3	0	2	0
		EW-WFD	90th%tile	15	0	0	10	3
		ASSETS	Annual Avg	20	5	0	2	1
		ASSETS	75th %tile	22	2	0	4	0
		ASSETS	90th%tile	18	2	0	5	3
	Discrete	EW-WFD	Annual Avg	16	2	4	3	2
		EW-WFD	75th %tile	17	2	3	0	5
		EW-WFD	90th%tile	11	1	2	5	8
		ASSETS	Annual Avg	14	4	3	2	4
		ASSETS	75th %tile	18	1	3	2	3
		ASSETS	90th%tile	14	3	0	2	8
Dissolved Oxygen WFD Framework 10th %tile Hourly Running Average of Annual Data	Hourly Running Average	EW-WFD	5th %tile	20	0	0	8	0
		EW-WFD	15th %tile	22	6	0	0	0
		ASSETS	5th %tile	5	9	14	0	0
		ASSETS	10 %tile	7	2	8	8	3
		ASSETS	15 %tile	18	6	0	4	0
	Instant-aneous	EW-WFD	5th %tile	20	0	0	8	0
		EW-WFD	10 %tile	28	0	0	0	0
		EW-WFD	15 %tile	23	5	0	0	0
		ASSETS	5th %tile	17	0	0	9	2
		ASSETS	10 %tile	18	4	0	6	0
		ASSETS	15 %tile	18	6	0	4	0

Table II-9. Sensitivity of ecological condition category derived from macroalgae biomass and cover to changes in data format and data integration.

Estuarine Class	Inlet Status	Segment	Dry Biomass					Wet Biomass				
			Annual Average/ Transect Average	Peak Season Average/ Transect Average	Peak Season Average/ Maximum Transect	Maximum Period/ Transect Average	Maximum Period/ Maximum Transect	Annual Average/ Transect Average	Peak Season Average/ Transect Average	Peak Season Average/ Maximum Transect	Maximum Period/ Transect Average	Maximum Period/ Maximum Transect
Enclosed Bay	Perennial	AHL	high	high	moderate	high	moderate	high	high	moderate	high	moderate
		BCF	high	moderate	very low	moderate	very low	high	high	very low	moderate	very low
		BQL	high	moderate	moderate	moderate	low	high	moderate	moderate	moderate	low
		MB	moderate	moderate	moderate	moderate	low	moderate	moderate	moderate	moderate	low
		SBF	high	moderate	moderate	moderate	moderate	high	moderate	moderate	moderate	moderate
		SDF	moderate	moderate	low	moderate	low	moderate	moderate	low	moderate	low
	Perennial-muted	BCM	low	low	very low	low	very low	low	very low	very low	very low	very low
		SBM	high	high	moderate	moderate	low	high	high	low	moderate	low
		SDM	moderate	very low	very low	very low	very low	moderate	low	very low	very low	very low
Lagoon	Ephemeral	SMC	low	very low	very low	very low	very low	moderate	low	low	low	very low
		UCL	very low	very low	very low	very low	very low	very low	very low	very low	very low	very low
	Intermittent	DL	very low	very low	very low	very low	very low	moderate	low	very low	very low	very low
		LPL	moderate	moderate	moderate	moderate	low	moderate	moderate	moderate	moderate	low
		SEL	high	moderate	moderate	moderate	moderate	high	moderate	moderate	moderate	moderate
	Perennial	GS	moderate	low	very low	low	very low	moderate	low	very low	low	very low
		MLF	low	very low	very low	very low	very low	low	very low	very low	very low	very low
		TJE	high	moderate	moderate	moderate	low	high	moderate	low	moderate	low
	Perennial-muted	BL	low	low	very low	low	very low	moderate	low	very low	low	very low
		BW	high	high	low	moderate	very low	high	moderate	low	moderate	very low
		MLM	very low	very low	very low	very low	very low	very low	very low	very low	very low	very low
		SAR	moderate	moderate	low	moderate	low	moderate	moderate	low	moderate	low
River Mouth	Intermittent	SCR	very high	very high	very high	very high	very high	very high	very high	very high	very high	very high
		SJC	high	very high	moderate	moderate	low	high	very high	high	high	moderate
		SME	moderate	low	very low	very low	very low	low	very low	very low	very low	very low
		TC	very high	very high	very high	high	high	very high	very high	very high	high	high
		ZC	high	very low	very low	very low	very low	high	low	low	low	very low
	Perennial	SDR	moderate	moderate	low	moderate	low	moderate	moderate	low	moderate	very low

Table II-10. Sensitivity of ecological condition category derived from phytoplankton biomass to changes in data format and data integration.

Class	Inlet Status	Code	Daily Average Data						Instantaneous Data						Discrete Data					
			IFREMER			ASSETS			IFREMER			ASSETS			IFREMER			ASSETS		
			Avg	75th %tile	90th %tile	Avg	75th% tile	90th %tile	Avg	75th %tile	90th% tile	Avg	75th %tile	90th %tile	Avg	75th% tile	90th %tile	Avg	75th %tile	90th% tile
EB	P	AHL	mod	mod	low	mod	mod	low	mod	mod	low	mod	low	low	high	high	mod	mod	mod	mod
		BCF	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high
		BQL	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	high	v.high	v.high	mod
		MB	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	high	v.high	v.high	mod
		SBF	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high
		SDF	v.high	high	high	v.high	mod	mod	high	v.high	high	mod	v.high	mod	v.high	mod	low	v.high	mod	mod
	P-M	BCM	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	mod	v.high	v.high	mod
		SBM	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high
		SDM	low	v.low	v.low	low	low	low	low	low	v.low	mod	low	low	low	low	v.low	mod	low	low
L	E	SMC	mod	high	low	mod	mod	mod	mod	mod	low	mod	high	mod	mod	mod	low	mod	mod	mod
		UCL	low	low	v.low	mod	mod	low	low	low	v.low	mod	low	low	mod	v.high	low	mod	v.high	low
	I	DL	high	mod	low	mod	mod	mod	high	mod	low	mod	mod	mod	high	low	low	mod	mod	mod
		LPL	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high
		SEL	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	low	mod	v.low	mod	mod	low
	P	GS	high	high	low	mod	mod	mod	high	mod	low	mod	high	mod	mod	low	low	mod	mod	mod
		MLF	mod	mod	low	mod	mod	mod	mod	mod	low	mod	mod	mod	v.high	high	high	v.high	mod	mod
		THE	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high
	P-M	BL	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high
		BW	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	high	v.high	low	mod	v.high	mod
		MLM	low	v.low	v.low	low	low	v.low	low	low	v.low	low	v.low	v.low	v.high	v.high	high	v.high	v.high	mod
		SAR	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high	v.high
RM	I	SCR1	v.low	v.low	v.low	low	low	low	v.low	low	v.low	low	v.low	low	v.low	v.low	v.low	v.low	v.low	v.low
		SCR2	v.low	v.low	v.low	low	low	low	v.low	low	v.low	low	v.low	low						
		SJC	low	v.low	v.low	low	low	v.low	low	low	v.low	low	v.low	v.low	v.high	v.high	high	v.high	v.high	mod
		SMR	low	low	low	mod	mod	low	low	mod	low	mod	mod	low	low	low	v.low	mod	low	low
		TC	v.high	v.high	high	v.high	v.high	mod	v.high	v.high	high	v.high	v.high	mod	high	v.high	low	mod	v.high	mod
		ZC	v.high	v.high	mod	v.high	v.high	mod	v.high	v.high	mod	v.high	v.high	mod	low	low	v.low	mod	low	low
	P	SDR	low	low	low	mod	mod	low	low	low	v.low	low	low	low	high	low	low	mod	mod	mod

Classes: EB- Enclosed Bay, L- Lagoon, RM- River Mouth
Inlet Status: P- Perennial, P-M- Perennial; Antrhopogenically Muted, E- Ephemeral, I- Intermittent

Table II-11. Sensitivity of ecological condition category derived from phytoplankton biomass to changes in threshold, data format, and data integration.

Class	Inlet Status	Estuary Code	Hourly smoothed						Instantaneous					
			EU-WFD			ASSETS			EU-WFD			ASSETS		
			5th %tile	10th %tile	15th %tile	5th %tile	10th %tile	15th %tile	5th %tile	10th %tile	15th %tile	5th %tile	10th %tile	15th %tile
Enclosed Bay	Perennial	AHL	moderate	high	high	moderate	moderate	very high	moderate	high	high	moderate	moderate	moderate
		BCF	high	high	high	very high	moderate	very high	high	high	high	moderate	very high	very high
		BQL	high	very high	very high	very high	moderate	very high	high	very high	very high	very high	very high	very high
		MB	very low	very low	very low	very low	moderate	moderate	very low	very low	very low	very low	very low	very low
		SBF	moderate	moderate	moderate	moderate	moderate	very high	moderate	moderate	moderate	moderate	moderate	moderate
		SDF	high	high	high	moderate	moderate	very high	high	high	high	moderate	moderate	moderate
	Perennial-muted	BCM	moderate	moderate	moderate	moderate	moderate	very high	moderate	moderate	moderate	moderate	moderate	moderate
		SBM	high	high	very high	very high	moderate	very high	high	high	very high	moderate	very high	very high
		SDM	high	high	high	very high	moderate	very high	high	high	high	moderate	moderate	very high
Lagoon	Ephemeral	SMC	very high	very high	very high	very high	moderate	very high	very high	very high	very high	very high	very high	very high
		UCL	very low	very low	very low	very low	moderate	very high	very low	very low	very low	very low	very low	very low
	Intermittent	DL	very low	very low	very low	very low	moderate	moderate	very low	very low	very low	very low	very low	very low
		LPL	moderate	high	high	moderate	moderate	very high	moderate	high	high	moderate	moderate	moderate
		SEL	low	low	moderate	moderate	moderate	very high	very low	low	moderate	very low	moderate	moderate
	Perennial	GS	very low	very low	very low	very low	moderate	moderate	very low	very low	very low	very low	very low	very low
		MLF	very high	very high	very high	very high	moderate	very high	very high	very high	very high	very high	very high	very high
		TJ	very low	very low	very low	very low	moderate	moderate	very low	very low	very low	very low	very low	very low
	Perennial-muted	BL	moderate	moderate	high	moderate	moderate	very high	moderate	moderate	high	moderate	moderate	moderate
		BW	very low	very low	very low	very low	very low	very low	very low	very low	very low	very low	very low	very low
		MLM	very low	very low	low	very low	moderate	very high	very low	very low	very low	very low	very low	very low
		SAR	moderate	high	high	moderate	moderate	very high	moderate	high	high	moderate	moderate	moderate
River Mouth	Intermittent	SCR1	very low	very low	very low	very low	moderate	very high	very low	very low	very low	very low	very low	very low
		SCR2	very low	very low	very low	very low	moderate	very high	very low	very low	very low	very low	very low	very low
		SJC	moderate	high	high	very high	moderate	very high	moderate	high	high	moderate	moderate	very high
		SMR	moderate	moderate	high	moderate	moderate	very high	moderate	moderate	high	moderate	moderate	moderate
		TC	low	moderate	moderate	moderate	moderate	very high	low	moderate	moderate	very low	moderate	moderate
		ZC	very low	low	moderate	moderate	moderate	very high	very low	low	moderate	very low	moderate	moderate
	Perennial	SDR	very low	very low	very low	very low	very low	very low	very low	very low	very low	very low	very low	very low

With respect to phytoplankton biomass, results were most sensitive to temporal integration of the data as well as thresholds (Table II-10). The difference between using daily averages versus the instantaneous data set did not have a large effect on the outcome of the assessment. However, there was a noticeable effect of using discrete data versus continuous data: 22% of systems increased in ecological condition class and 19% decreased in condition class when discrete data is used compared to continuous daily averages. Changing the data integration period also has a significant effect on the outcome. Using a 75th or 90th percentile instead of the annual average resulted in 22 and 44% of segments that change ecological condition class, respectively. Changing the assessment framework also had an effect on the assessment outcome. Using ASSETS as described (90th percentile), 7% of segments increased in score and 30% decreased in score relative to IFREMER thresholds applied to annually averaged data.

DO assessments were sensitive to changes in temporal integration and assessment framework. Use of the 5th percentile resulted in 8 segments scoring lower, whereas use of the 15th percentile resulted in 22% of segments scoring higher in ecological condition than the 10th percentile (Table II-11). Applying ASSETS thresholds to the 10th percentile of continuous data resulted in category change in 78% of segments, with roughly equal numbers of segments increasing and decreasing in score. Changing data format from an hourly running average to instantaneous generally had no effect on results, with the exception of the ASSETS framework, in which 37% of segments changed class when discrete data were used.

Discussion

Extent and Magnitude of Eutrophication in Southern California Bight Estuaries

According to the EU-WFD, the Bight’08 assessment found that eutrophication is pervasive in SCB estuarine segments based on indicators and thresholds developed for the EU-WFD. The percentage of systems characterized as having moderate or worse ecological condition varied by indicator, from 78% for macroalgae, 37% for phytoplankton and 59% for dissolved oxygen. Using the indicators as part of a multiple lines of evidence approach provides different answers, but does not change the answer that eutrophication is pervasive in the segments monitored. EU-WFD applies a "one out, all out" approach in determining ecological status wherein the lowest score for any single element becomes the overall score for the state of the waterbody (Borja *et al.* 2004, Zaldivar *et al.* 2008). Applying this to the Bight’08 assessment, all but one of the segments assessed would require management action to improve ecological condition. However, several studies have demonstrated the short comings of using a single indicator to establish ecological condition and that use of multiple metrics provide a more robust accounting of condition (Borja *et al.* 2009a, Borja *et al.* 2009b, Borja and Rodriguez 2010, Borja *et al.* 2011). The ASSETS framework utilizes multiple lines of evidence where in each indicator is scored based on intensity of expression with respect to threshold values, spatial extent and frequency of occurrence. Scores for primary symptoms (primary producer response) and secondary symptoms (dissolved oxygen) are combined to generate an overall score of ecological condition for the estuary (Bricker *et al.* 2003). The applicable "primary symptom" would vary depending on the segment in the SCB. For example, SCR and SJC had little macroalgae (categorized as "very high ecological condition" based on macroalgae biomass and cover), but high suspended chlorophyll *a* (categorized as "very low" for SCR and "low" for

SJC), indicating that eutrophic condition in these systems is driven by phytoplankton response instead of macroalgal response. This underlies the importance of selecting the most critical primary producer response indicator for each system, rather than a one size fits all approach (Bricker *et al.* 2003, Zaldivar *et al.* 2008, Borja *et al.* 2009b). A variation on the ASSETS primary and secondary symptoms strategy could be applied to the Bight'08 assessment by requiring that each segment must score moderate or worse based on one of the two primary producer indicators because they would not necessarily co-dominate) and dissolved oxygen. This would result in 53% of segments requiring management action to improve ecological condition. Thus, based on the most conservative estimate of eutrophication in each estuary, over half of systems surveyed would require some management action according to a multi-metric application of the EU-WFD framework.

We qualify these results by acknowledging that, as a regional survey rather than a site-specific estuarine assessment, the results are based on data collected in 1-2 segments per estuary and were not intended to characterize the entire estuary. However, in roughly half of the estuaries, the segment represents 75% or more of the total estuarine area because SCB estuaries are typically small. The segments were selected to be proximal to the source of freshwater nutrient loading rather than the ocean inlet and, thus, represent a location within the estuary that is more likely to have a problem with eutrophication. However, our assessment was conducted over a one year period, rather than a one-time site visit that is typical of regional or national surveys (Nelson *et al.* 2005). Therefore, we feel confident that we captured seasonal variability in indicator expression fairly well. However, we acknowledge that inter-annual variability in nutrient loading will greatly affect expression of eutrophication symptoms and we acknowledge this limitation in our estimates of eutrophication. Finally, spatial variability within the segment was fairly well documented with macroalgae, but monitored only at single point in the bottom waters for dissolved oxygen and phytoplankton. The implications of these decisions on the assessment results and recommendations for future development of assessment frameworks and subsequent regional assessments are explored in the sections below.

SCB Estuaries Compared to Other Estuaries

Widespread coastal eutrophication has been reported for estuaries in the United States in the National Estuarine Eutrophication Assessment (NEEA), but the status of southern California estuaries was largely unknown (Bricker *et al.* 1999, Bricker *et al.* 2008). The majority of NEEA assessed estuaries showed signs of eutrophication, representing 78% of assessed estuarine area, falling into moderate or worse ecological condition conditions according to the ASSETS assessment framework and symptoms of eutrophication were predicted to worsen by 2020 in 65% of estuaries and improve in 20% of estuaries. The majority of US estuaries assessed (65%) displayed at least one symptom of eutrophication, suggesting a large-scale, national problem. Of the systems assessed, 29% had low or very low ecological condition and were characterized by symptoms that are extensive (covering 50% or more of the system) and/or are persistent. Though the largest proportion of the estuaries (40%) surveyed had moderate eutrophic condition ratings characterized by symptoms that are periodic and occur over a moderate proportion of the estuary. A study of eutrophication throughout the European and Australian coastlines found that symptoms of eutrophication were also prevalent, using the ASSETS assessment framework (Hillman *et al.* 1990, Ærtebjerg *et al.* 2001, Borja *et al.* 2004, Ferreira *et al.* 2007, Borja *et al.* 2009a).

Other studies which employ ASSETS for making an assessment of eutrophication found various expressions of eutrophication regionally, In 14 Basque Country (northern Spain) estuaries indicated that all systems fell in a moderate or worse ecological condition category using the ASSETS framework (Garmendia *et al.* 2012). In Portugal, 3 out of 7 estuaries were in a moderate ecological condition category and 4 in a high or very high category, no estuaries fell in a low or very low category (Whitall *et al.* 2007b).

Estuaries with high overall eutrophic conditions in the NEEA study were generally those that received the greatest watershed nutrient loads (Bricker *et al.* 1999). Similarly, macroalgae and phytoplankton response in SCB estuaries was also found to be significantly correlated with watershed nutrient loads (Section V), and dissolved oxygen was found to be significantly correlated with sediment organic matter (Section IV). Watershed nutrient loads into estuaries assessed in the NEEA study were generally 75% higher compared to loads into SCB estuaries overall, and nutrient loads into the European estuaries were twice as high as loads into the SCB, but the some of the highest loads from individual watersheds into the SCB are comparable to the highest loads in other regions (Sengupta *et al.* submitted). SCB estuaries have a similar distribution between moderate to very low ecological condition as estuaries in the NEEA and European studies. Though it should be noted that the NEEA survey and the European studies were not based on a probabilistic design and include systems for which there was existing data. Given that loads into the SCB are lower but the ecosystem response it similar, it seems that many SCB estuaries are more susceptible to eutrophication compared to estuaries in other parts of the US due to restricted hydrology and/or other factors known to effect estuarine response to eutrophication (Pinckney *et al.* 2001, Zaldivar *et al.* 2008).

Appropriateness of Indicators and Thresholds for SCB Estuaries

SCB estuaries incorporate a wide range of habitat types and thus are host to a variety of species which could potentially be used either as an indicator of eutrophication directly or to inform appropriate thresholds for indicators. We chose to make our assessment using three common indicators of eutrophication; however, the relevance of the indicators and assessment frameworks as well as whether the sampling approach adequately captured the status of those indicators in different settings in SCB estuaries is worth exploring. It is worthwhile to acknowledge upfront that the authors of the ASSETS (Bricker *et al.* 2003) and EU WFD frameworks (Scanlan and Wilson 1999, Souchu *et al.* 2000, Best *et al.* 2007, Zaldivar *et al.* 2008) recognize that the lack of data both on ecosystem response to nutrient over enrichment as well as on reference condition may mean that the applicability of indicators to specific habitat types, the thresholds as well as how event duration and frequency is incorporated is likely to change over time as the body of literature on eutrophication grows (Patricio *et al.* 2007, Scanlan *et al.* 2007, Domingues *et al.* 2008). Our intent is to inform this debate by discussing to what degree these frameworks are applicable to SCB estuaries.

Macroalgae. While macroalgae is a natural component of estuaries, an overabundance of macroalgae can have adverse effects on estuarine ecological health, including altered sediment and water column chemistry (e.g., hypoxia, ammonia, and sulfide toxicity), smothering of seagrass and reef habitats, and adverse effects to benthic invertebrates on intertidal flats and subtidal habitat (Soulsby *et al.* 1982, Fletcher 1996, Raffaelli *et al.* 1999, Green 2011). For these reasons, macroalgae has been identified as a relevant indicator to assess eutrophication in estuarine environments (Bricker *et al.* 2003, Scanlan *et al.* 2007), particularly in California's bar-built lagoon and river mouth estuaries that are dominated by shallow subtidal or intertidal habitat (Sutula 2011).

Macroalgae was the dominant aquatic primary producer in SCB estuaries (Section III) and was applicable as an indicator for most segments. The Scanlan *et al* (2007) macroalgal assessment framework used to assess ecological condition worked well, as it accounted for both the abundance and spatial patchiness inherent in this indicator. Measurement of macroalgae on intertidal flats may be less relevant in seagrass-dominated estuaries if preservation of seagrass communities is the desired end-point (MB, SDF, BQL, AHL, SBF, SBM). In this case, the assessment of macroalgal effects are better made in the seagrass beds (Huntington and Boyer 2008). In addition, while our monitoring approach worked well in sites with large intertidal flats, it was less effective for assessment of biomass and cover of floating mats typically found in estuaries with "closed" tidal inlets (TC, SMC, UCL, DL, SJC, SCR). These estuaries have little to no intertidal area (due to closure of the ocean inlet) and thus, the floating algal mats do not become entrained on intertidal flats, but are distributed, often unevenly, throughout open water area. Our protocol involved returning to a fixed transect, which often results in over- or under-estimation of biomass in these systems as wind often tends to push the algae to one side of an estuary (e.g., ZC, SMC). Further work is required to develop effective protocols to capture areal extent and biomass of floating macroalgal mats in systems with a closed tidal inlet.

We chose not to modify the EU-WFD proposed thresholds for macroalgae, but it is certainly important to consider their relevance for SCB estuaries. Scanlan *et al*. (2007) proposed "actionable" levels of macroalgae at > 70 g dw m^{-2} and > 25% cover, or > 130 g dw m^{-2} and > 5 % cover. Results of a recent study by Green *et al*. (submitted) in Bodega Bay and Newport Bay show elimination of surface deposit feeders, an important functional group of invertebrates for fish and bird foraging, after four weeks at 110-120 g dw m^{-2} and 100% cover. Similarly, Bona (Bona 2006) showed a clear effect threshold on benthic habitat quality at > 700 g ww m^{-2} (90 g dw m^{-2}) and > 70 % cover. An experiment by Green (2011) in Mugu Lagoon showed that treatments of 75 g dw m^{-2} and 100% cover did not experience significant declines in benthic infaunal communities after 8 weeks of exposure, but this estuary is acknowledged to be highly eutrophic with year-round algal blooms where infaunal communities were likely already stressed. Therefore, an "effects" threshold in the range of 70 - 120 g dw m^{-2} (as described in (Scanlan *et al.* 2007)) is reasonable. At what aerial percent cover this threshold is applied is another question. Scanlan *et al* (2007) uses cover categories of <5%, 5-15%, 15-25%, 25-75% and > 75%. It was our experience that there was not much difference in the ranking of estuaries between 5-15% versus 15-25%, while a number of estuaries fell within the larger 25-75% range. In SCB segments, placement of 10 of segments (37%) in the "moderate" ecological condition was largely driven by cover greater than 25% but less than 50% and biomass less than 70 g dw m^{-2}. Placement of these segments (BCF, BQL, LPL, MB, SAR, SBF, SDF, SDR, SEL) in an "actionable" category may be overly conservative. It may be sensible to consider refinement (collapsing 5-25%) and adding cover categories between 25 and 75% cover.

Duration was not explicitly assessed in Scanlan *et al*. (Scanlan *et al.* 2007) but was incorporated into this assessment by taking the average of the two consecutive peak periods, equivalent of having a bloom duration of a minimum of 8 weeks. Green (2011) and Green *et al*. (submitted) found that the rapidity of decline in benthic infaunal communities strongly depended on the amount of biomass, thus demonstrating the importance of including duration in combination with biomass (magnitude) and cover (extent). They observed that continuous macroalgae coverage by mats with a thickness of 120-186 gm dw m^{-2} for 4-8 weeks would show declines in surface deposit feeders. For the SCB segments sampled, 37% of segments (BCM, BL, DL, GS, MLM, SAR, SDF, SDM, SDR, UCL) had moderate or worse biomass for 12 or more weeks (3 or more consecutive periods) and 26% (BCM, BL, DL, GS, MLM, SDR, UCL) had moderate or worse biomass for longer than 20 weeks (5 or more consecutive periods). Thus, it is reasonable to expect that at 25% or more SCB segments have sufficient bloom duration to significantly affect benthic infaunal communities.

Data management also had a significant impact on how sites were scored. Data management decisions for macroalgae include whether to use wet or dry biomass, whether to use the mean biomass from the three transects, the maximum biomass, or a percentile, and the time period of data integration. Macroalgae biomass was measured in terms of both wet and dry weights. The EU-WFD uses thresholds based on wet weights for practical reasons (Scanlan *et al.* 2007); although, recent work has argued for use of dry weights due to the unequal water weight retained by different samples (Patricio *et al.* 2007). We observed that wet weights and dry weights were not necessarily linearly related, with significant scatter particularly for higher biomass samples ($r^2 = 0.691$, $p < 0.0001$). Thus, we felt that dry biomass was a more scientifically defensible approach to assessment of eutrophication. That said, using our preferred data integration period (average biomass and cover from two consecutive periods of highest biomass and cover), while 8 segments changed category, only 2 segments changed crossed the moderate/high threshold, the critical threshold for management action (one improving, BCF, and one declining, BW). Management of spatial data also had an impact on score. We utilized average biomass and cover from all three transects in an effort to weight all intertidal area in the segment equally and thus generate a score representative of the entire segment rather than the most severely affected sub-section. Use of a percentile or only using biomass and cover data from the worst of the three transects generated lower scores in almost half of the segments, demonstrating the importance of variation in spatial scales in assessment. More research is required to identify the amount of estuarine area that can be impacted by significant and sustained algal blooms before the ecosystem is significantly degraded. Finally, how temporal data is integrated also affects how estuaries are categorized. This final question would exist if duration is explicitly incorporated into the assessment framework as described above, but it was instructive to determine the effect of averaging biomass and cover data over different timescales. As expected more segments scored in higher ecological condition categories using an annual average versus peak season versus maximum period; what was interesting was the differences between peak season (average of two consecutive periods of high biomass and cover) and the maximum period. For some sites a maximum period with very high biomass and cover was averaged with a period of relatively low biomass and cover resulting in a moderate score. This approach defined the difference between sites with chronic problems and those with short-duration blooms.

Phytoplankton. Phytoplankton biomass, measured as water column chlorophyll *a*, is a common indicator of eutrophication in inland, estuarine and coastal waters (Cloern 2001, Bricker *et al.* 2003).

Elevated chlorophyll *a* concentrations reduce light to benthic primary producers, including seagrass and microphytobenthos, create conditions leading to hypoxia, and are associated with the promotion of harmful algal blooms (see (Sutula 2011) for comprehensive review). Phytoplankton biomass is most applicable in estuaries that are dominated by sub-tidal habitat with longer residence times (Nielsen *et al.* 2004) and not relevant intertidally-dominated estuaries (BW, LPL, MLM, MLF, SEL, SJC, TJE). In shallow subtidally-dominated estuaries, it is difficult to predict how a given estuary will respond to nutrient loading with regard to which primary producer group will dominate (Short *et al.* 1995, Flindt *et al.* 1999); some systems will become dominated by macroalgae and others by phytoplankton. The dominant primary producer group in perennially tidal estuaries may be related to water residence time, because low residence time favors macroalgae while longer residence time favors phytoplankton (Valiela *et al.* 1997). In intermittently tidal estuaries during a closed inlet condition, no work has been done to help predict when macroalgae will dominate over phytoplankton.

Another consideration is whether existing thresholds for phytoplankton are appropriate for SCB estuaries. Both IFREMER (Souchu *et al.* 2000) and ASSETS (Bricker *et al.* 2003) establish thresholds based on the paradigm of light limitation of benthic primary producers, in particular seagrass, although references to other adverse effects are made. Using ASSETS rather than IFREMER on annually averaged data resulted in an upgrade in condition class of approximately 25% (Table II-7). Both ASSETS and IFREMER assessment frameworks have similar "no effect" levels of <5 to 7 µg L^{-1}. Moderate effects range from roughly 7 to 10 µg L^{-1}; similar to the chlorophyll *a* guidance or criteria established for Yaquina Bay (3-5 µg L^{-1}; Brown *et al.* 2007) and Florida estuaries (<3.8-11.0 µg L^{-1}; Janicki *et al.* 2000, Janicki *et al.* 2009). Above 20 µg L^{-1}, submerged aquatic vegetation shows declines (Stevenson *et al.* 1993) and phytoplankton community shifts from diverse mixture to monoculture (Twilley 1985b). At 60 µg L^{-1} chlorophyll-a, high turbidity and low bottom water dissolved oxygen have been observed in estuaries (Jaworski 1981, Bricker *et al.* 2003). Because closed estuaries are typically brackish and can become dominated by cyanobacteria under high nutrient loading (Magrun 2011); studies of the relationships between chlorophyll *a* and cyanobacteria blooms in lakes can be illustrative (Walker 1985, TetraTech 2006). Cyanobacteria blooms will almost never occur when summer mean chlorophyll *a* concentrations are less than 5 µg L^{-1}, while concentrations of 10 µg L^{-1} would imply that such blooms are rare. These values are comparable to "no effect" levels in seagrass dominated habitats as described by ASSETS and the IFREMER. Similarly, concentrations of 20 µg L^{-1} suggests cyanobacteria blooms will occur about 15-20 percent of the time, which has been suggested to be the maximum allowable level consistent with full support of contact recreation use, and a mean concentration of 25 µg L^{-1} corresponds to blooms about 25% of the time (Walker 1985). Thus, while there are a few studies that provide a clear picture of dose-response for phytoplankton biomass, there appears to be some scientific consensus around ranges of thresholds. Additional work is needed to refine these thresholds for SCB estuaries.

Method of data collection and type of averaging applied to the data set had an impact on the condition categories of a significant number of segments. We collected both continuous and discrete chlorophyll *a* data, and while the discrete data was used to convert the continuous fluorescence data into concentrations, applying thresholds to the two data sets had a significant impact on the scores of some of the systems. Half of the systems changed condition category when the discrete data was used versus the continuous data and 25% of the systems crossed the high/moderate boundary indicating a change in whether management action would be taken. It is worth noting that our discrete data set was insufficient for use in the IFREMER (which requires data collection at minimum of every month). However, more research is required to determine how much discrete data would be required to make a reasonable assessment of eutrophication using phytoplankton chlorophyll *a* or whether continuous data must be collected. Continuous data could be expressed as either instantaneous 15 minute data or as daily averages. We opted to use daily averages to eliminate some high frequency noise in the data set because it is the sustained blooms that impact ecosystem health. However, comparison between the two data sets indicated that there was not a significant effect on how data were categorized with respect to ecological condition (one site, SDF, moved from "very high" to "high").

Similar to macroalgae, phytoplankton bloom duration should also be considered when assessing the impact of a bloom on estuarine ecological condition. Determining what level of chlorophyll *a* constitutes a bloom sufficient to impact ecosystem health varies based on phytoplankton species and environmental factors such as depth (Batiuk *et al.* 2001, Wazniak and Hall 2005, Wazniak *et al.* 2007). Phytoplankton blooms of short duration are vital to sustain estuarine food-webs (Cloern 1996, Cloern and Jassby 2008). However blooms lasting longer than 1 to 2 months will begin to have a negative impact on submerged aquatic vegetation, decreasing habitat diversity and impacting ecological condition (Moore and Wetzel 2000, Ruiz and Romero 2001). Within the SCB, 19% of segments had continuous phytoplankton greater than 10 $\mu g\ L^{-1}$ for longer than 2 months (MLM, SCR, SDM, SDR, UCL). For biomass greater than 30 $\mu g\ L^{-1}$, 15% of segments (MLM, SCR, SDM, UCL) exceeded this threshold continuously for 1 month and 7% for 2 months (SCR, SDM). Thus, it is reasonable to assume that up to 25% of SCB segments could not sustain healthy seagrass habitat with measured duration of phytoplankton bloom. However, only 25% of SCB estuaries had significant seagrass habitat, although all estuaries had benthic microalgae and 30% of the systems had brackish water submerged aquatic vegetation (*Ruppia spp. spp;* Section III) which could potentially be adversely affected by phytoplankton shading (Verhoeven 1980). Additional work is needed to investigate the effects of bloom duration on these habitats.

Dissolved Oxygen. DO is necessary to sustain the life of all aquatic organisms that depend on aerobic respiration. DO concentrations reflect an equilibrium between oxygen-producing processes (e.g., photosynthesis) and oxygen-consuming processes (*e.g.,* respiration), and the rates at which DO is added and removed from the system by atmospheric exchange (aeration and degassing) and hydrodynamic processes (e.g., accrual/addition from rivers and tides vs. export to ocean) (Diaz 2001, Caffrey 2004). Dissolved oxygen can be mixed into the bottom waters where it can support the life of epibenthic organisms. Oxygen diffuses into sediments or is advected in through the actions of benthic infauna (bioirrigation or bioturbation) and tidal pumping. Eutrophication produces excess organic matter that fuels the development of hypoxia and, in some cases, anoxia (<0.5 mg DO L^{-1}) as that organic matter is respired (Diaz 2001). Thus low DO has a direct linkage to aquatic life and beneficial use protection (See Sutula *et al*. 2012 for comprehensive review). Like phytoplankton biomass, DO is most applicable in subtidal habitats; application in intertidal habitats is problematic as mudflat and marsh have an abundance of dissolved organic carbon sources which consume oxygen and lead to lower expected dissolved oxygen levels in back basins and tidal channels. However, no guidance is available to determine at what point DO should no longer be applied as an indicator.

ASSETS (Bricker *et al.* 2003) and the EU-WFD for DO (Best *et al.* 2007) thresholds are based on observed impacts of hypoxia on benthic and demersal fauna as well as expert opinion and are targeted to be relevant in a wide range of estuarine environments (Vaquer-Sunyer and Duarte 2008; Table II-6). Our use of the EU-WFD framework in SCB estuaries was well-grounded. The thresholds proposed by Best *et al*. (2007) are similar to those calculated for California species (5.7 mg L^{-1} as chronic-effects criteria protective of 95% of the non-salmonid population and 2.8 mg L^{-1} as acute effects criteria; Sutula *et al*. 2012). For salmonids, Sutula *et al*. (2012) calculated 6.3 mg L^{-1} as chronic effects criteria and 4.0 mg L^{-1} as acute effects criteria, but notes that the effects data used to calculate these criteria were based on freshwater exposure studies. The Best *et al*. (2007) thresholds have the advantage of incorporating the salinity effects on oxygen solubility, and thus, can reconcile a threshold protective of all life history stages for salmonids from 7 mg L^{-1} in freshwater to 5.7 mg L^{-1} at marine salinities. The ASSETS upper threshold of 5.0 mg L^{-1} is roughly equivalent to this threshold but does not take into account salinity (Bricker *et al.* 2003). Thus, applying ASSETS to river mouth and lagoonal estuaries with a closed inlet, habitats that are typically brackish and that currently or historically support salmonids in southern California, could be under protective. In addition, Sheldon and Alber (2010) revealed some confusion in the literature over the definition of hypoxia, often cited as < 2 mg L^{-1}, but the units used to describe oxygen concentrations cited criterion for hypoxia of 2 ml O2 L^{-1} (Diaz and Rosenberg 1995) is actually equivalent to approximately 2.8 mg O_2 L^{-1}, which is equivalent to the acute criteria calculated by Sutula *et al*. (2012). The use of the Best *et al*. (2007) versus ASSETS thresholds has a large effect on categorization. Relative to Best *et al*. (2007), 78% of segments changed class using ASSETS, with 40% changing two or more condition categories.

The response of aquatic organisms to low dissolved oxygen will depend on the intensity of hypoxia, duration of exposure, and the periodicity and frequency of exposure (Rabalais and Harper 1992). Both ASSETS and EU-WFD (Bricker *et al.* 2003, 2007) utilize a percentile approach to data integration, calculated by ranking the continuous data from lowest to highest value, and applying the percentile. The EU-WFD applies a fifth percentile and ASSETS a tenth percentile; thus the 5th percentile of 9 months of continuous DO data equates to approximately 2 weeks below a designated threshold. Use of 5th and 15th percentile relative to 10% changes down- and up-grades condition classes in 20-30% of segments. The use of the percentile approach integrates the duration and frequency of low DO events and doesn't distinguish between high frequency short duration events and low frequency but long duration events. The effect of these two examples can be very different on biota, depending the timing and number of reproductive cycles in the year, number per brood, etc. Sutula *et al*. (2012) also note that natural hypoxia in bottom waters of bar-built estuaries is an issue for application of DO thresholds and one which certainly effects the interpretation of the results of this assessment. Shallow estuaries are prone to development of density-driven stratification during restrictions or closure to tidal exchange when the estuaries stratify with dense, salty waters on the bottom and fresher water at the surface thus precluding diffusion and mixing of oxygen to bottom waters (Largier *et al.* 1991, 1996). All of the estuaries that were closed to tidal exchange in this assessment were typified by hypoxic events greater than 1.5 days in duration, with some of the more eutrophic estuaries having hypoxic events up to 36 days (Section IV). Sutula *et al*. (2012) notes that studies of natural hypoxia in minimally disturbed "reference" estuaries are needed.

Notably, ASSETS goes one step further and includes a frequency category in addition to categories for magnitude and spatial extent. The EU-WFD also recommends using Fundamental Intermittent Standards (FIS) and a return period in more sensitive habitat that sets a minimum threshold for dissolved oxygen, a duration, and a return period. They proposed a second tier FIS for "high" ecological condition that dissolved oxygen should not fall below 2 mg L^{-1}, more frequently that once every 6 years over a 6 hour tidal cycle and for "moderate" status not more than once every 3 years (Best *et al.* 2007). Because we only have one year of data, we could not include this second tier standard.

Considerations for Future Assessments

Incorporating Spatial Variability. Southern California estuaries are highly variable: in size and aspect (form, sinuosity, width, etc.), hydrologic connectivity to the ocean, estuarine area versus watershed size, and other characteristics. A substantial fraction of these systems are small, shallow (<2 m deep), benthically-driven systems (e.g., Topanga Canyon Lagoon, Ballona Wetlands, etc.), while others are large, more pelagic systems (e.g., San Diego Bay, Mission Bay). Many are subjected to natural seasonal closure due to the development of a sandbar at the inlet, and consequently are subject to natural periods of hypoxia when the systems stratify following closure (Largier *et al.* 1991, Largier *et al.* 1996) and the spatial extent of "natural" hypoxic volume is unknown.

For the Bight'08 assessment we chose to monitor an index area in each system that would be most likely to exhibit symptoms of eutrophication. For some of the smaller systems, this targeted segment comprised the entire estuary, while for others it comprised a small fraction of the system where water residence time was likely to be longest (Table II-1). Consequently, spatial variability within an estuary was not addressed. Scanlan *et al*. (2007) recognized the problem of spatial variability in expression of macroalgae biomass and cover and the impact on assessment of ecological condition. In terms of spatial variation, they proposed using average biomass and cover values over the entire intertidal area, rather than using an index area comprised of only those flats where algal blooms occurred, recognizing that this would under-represent problems in these sub-areas. Our sensitivity analysis clearly indicates that spatial extent impacts ecological categorization in SCB estuaries (using a single transect with highest biomass and cover scores segments in a lower condition class compared to using the average of the three transects). However, the larger the monitoring area, the more resource intensive it would be to monitor. Thus, more research is needed to identify the optimal assessment area relative to the size of the estuary.

Best *et al*. (2007) also recognized that there were trade-offs between collecting continuous dissolved oxygen data in a relative few stations versus collecting discrete samples over a larger area. Continuous data allows for a confident assessment of length and frequency of hypoxic events, but deployment of *in situ* monitors at multiple stations is resource intensive and too few monitoring locations may not reflect the general condition of the estuary. Frequent spot measurements allows for a good estimation of extent but no means of accurately assessing event duration and frequency (Best *et al*. 2007). For our study we measured dissolved oxygen in bottom water at a single location in each segment where water residence time was expected to be longest, thus generating a very conservative estimate of frequency and duration of hypoxia in each segment. More research is required to investigate how many monitoring stations would be needed to adequately assess the hypoxic volume of each estuary, as well as a means to collect data on stratification (surface dissolved oxygen versus bottom water dissolved oxygen). Furthermore, more research is needed to determine if thresholds should vary by estuarine class and whether systems that experience "natural" hypoxia due to closure of the inlet should have different thresholds compared to more well-flushed systems.

Incorporating Temporal Variability. Seasonal variability must be taken into account when developing an appropriate monitoring program. Devlin *et al*. (Devlin *et al.* 2011) showed that use of annual data and sampling throughout the year, with inclusion of frequency of occurrence, provided a more representative assessment of trophic status in estuarine case studies. In a study in French Mediterranean lagoons, different results could be obtained as a function of the sampling frequency and sampling season (Zaldivar *et al.* 2008). In the SCB, timing of peak macroalgal and phytoplankton abundance varies widely across estuaries (Section III). Most systems have peak biomass in the spring through fall (with some studies having a peak in spring and another peak in fall, which is common in estuaries (Scanlan *et al.* 2007)); however a significant fraction had peak biomass during the late fall and winter months (Section III). Furthermore, the time of year for most frequent hypoxic events or events of long duration is also seasonally variable (Section IV). Estuaries with seasonal restriction/ closure of the ocean inlet are most susceptible to hypoxia when tidal flushing is reduced (Largier *et al.* 1991, 1996, 1997). However, SCB estuaries experience this restriction at different times of the year; with some systems are prone to closure during the winter (e.g., Los Penasquitos Lagoon) and some during the summer (e.g., Topanga Canyon), and consequently would be most sensitive to hypoxia at different times (Section IV). Thus, seasonal variability presents a challenge for developing a monitoring program to assess eutrophication in SCB estuaries. Many SCB estuaries spend as much time in a "high" or "very high" ecological condition category as they spend in a "moderate" or worse category, and results of the sensitivity analysis indicate that how temporal data is integrated has a significant effect on the ecological condition score. Our study characterized seasonal variability reasonably well through a combination of continuous *in situ* monitors and every other month sampling. However, such a monitoring program would be costly to maintain over the long term. Consequently, *in situ* monitors should be deployed continuously over an annual cycle and discrete sampling of primary producer biomass would have to occur several times a year until the critical periods for hypoxia and blooms could be identified for each estuary at which point the monitoring program could potentially be modified to cover only the critical period for specific systems.

Changes in nutrient and organic matter loading into southern California estuaries can differ dramatically from year to year (Sengupta *et al*. submitted) and this temporal variability must also be taken into account when assessing ecological condition in estuarine environments. Inter-annual variability has been well documented for macroalgae spatial coverage and biomass (Raffaelli *et al.* 1999, Martins *et al.* 2001, Patricio *et al.* 2007), phytoplankton blooms (Peterson *et al.* 1985, Jassby and Powell 1994, Perez-

Ruzafa *et al.* 2005), and hypoxia (Diaz and Rosenberg 1995, Kemp *et al.* 2005, Breitburg *et al.* 2009) in a variety of estuarine settings. Nitrogen and phosphorus loading to SCB estuaries during the 2008-2009 water year was moderate compared to annual loads over the past 13 years (Sengupta *et al*. submitted), so we expect that our data is fairly representative of the average condition. However, we recognize that a single year of data could be misleading if other factors result in a higher or lower response in any of the indicators. Thus, several years of monitoring data should be collected before making an overall assessment of any given system. Indeed, the proposed EU-WFD frameworks for each indicators recommends monitoring be conducted in at least 3 out of the 5 year reporting cycle (Best *et al.* 2007, Scanlan *et al.* 2007, Zaldivar *et al.* 2008). Though how many years of monitoring are required to make an accurate assessment of ecological condition in SCB estuaries is an open question.

Multiple Lines of Evidence. Whether to utilize multiple lines of evidence and how indicators should be integrated into an overall assessment for SCB estuaries will require further investigation. The ASSETS framework fully integrates biological response indicators with indicators of pressure (e.g., nutrient loading) as well as future outlook (Bricker *et al.* 2003). A similar approach could be adopted for SCB estuaries, though how to integrate the pressure and state metrics would require further exploration. Primary production in SCB segments was significantly correlated with present-day nutrient loading, whereas dissolved oxygen was only significantly correlated with sediment organic matter. Thus, each indicator responses to nutrient loads on different time scales and could thus generate different results for ecological condition (estuaries going from high ecological condition to low may score lower on primary producer indicators than dissolved oxygen indicators, whereas estuaries improving in ecological condition may score lower on dissolved oxygen than primary producers). Furthermore, which indicators should be required in a multi-metric approach should also be investigated. This study included assessments of two primary producer groups; however, research has shown that phytoplankton and macroalgae do not necessarily co-dominate in eutrophic estuaries and an overgrowth of one group may prohibit growth of another (Taylor *et al.* 1995, Zaldivar *et al.* 2009). Consequently, the two indicators may generate conflicting results for ecological condition score, as was the case for some SCB estuaries, notably SCR and SJC. Consequently, only one primary producer group should be included in a multiple lines of evidence approach.

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III. PATTERNS IN AQUATIC PRIMARY PRODUCERS IN SOUTHERN CALIFORNIA BIGHT ESTUARIES

Introduction

Estuarine aquatic primary producers, comprised of algae (phytoplankton, microphytobenthos, and macroalgae) and submerged aquatic vegetation, play a key role in estuarine function and ecosystems services, including the fundamental structuring of food webs and mediation of biogeochemical cycling, particularly nutrient and organic matter cycling (Kennish 1990, Sand-Jensen and Nielsen 2004). For example, the fate of aquatic primary producer carbon varies depending on the type of marine plant (Duarte and Cebrian 1996). Slow growing submerged aquatic vegetation decompose slowly and most of the carbon production is channeled directly to decomposers and a larger fraction is stored in the sediments whereas more than half of the carbon produced by fast growing macroalgae and phytoplankton is transferred to herbivores and is thus available to higher trophic levels (Duarte 1992).

The relative dominance of the four major types of aquatic primary producers -- phytoplankton, macroalgae, microphytobenthos, and submerged aquatic vegetation -- are controlled by a suite of a factors that vary with respect to the basic physiological requirements of each group and present environmental constraints to their stability, growth and reproduction (Day 1989). These factors include: 1) light, 2) water depth, 3) temperature, 4) desiccation, 5) water velocities and turbulence, 6) gradients in water column and sediment nutrient and organic matter availability, and 7) grazing by consumers. The interplay of these factors controls the presence and relative dominance and abundance of primary producer groups within estuarine subtidal and intertidal habitat types and across estuarine classes. These four primary producers tend to distribute themselves in predictable patterns across tidal inundation gradients found in estuaries. In turbid or deepwater subtidal habitats, particularly in wave dominated environments, phytoplankton tends to be the dominant primary producer, or co-dominant with microphytobenthos in deepwater habitats with high water clarity (Day 1989, Wetzel 2001). As depths decrease towards the shallow subtidal zone and particularly in estuaries with a strong tidal regime, microphytobenthos, submerged aquatic vegetation, and macroalgae that are attached to sediment are at a competitive advantage over phytoplankton, which can be easily flushed out during a tidal cycle (Figure III-1; Valiela *et al.* 1997). Depending on water residence time, nutrient availability, substrate, and other factors, phytoplankton, submerged aquatic vegetation, microphytobenthos, and macroalgae can co-dominate in shallow subtidal habitat (>10 m in depth). In intertidal flats, macroalgae and microphytobenthos are generally the dominant primary producers.

Understanding the basic spatial and temporal patterns of aquatic primary producer abundance, composition and the factors driving these patterns is fundamental to the sustainable management of estuarine resources. Data on the abundance (biomass) and relative composition of estuarine aquatic primary producer groups are typically used in the context of assessing the extent of eutrophication (Bricker *et al.* 2003, Zaldivar *et al.* 2008). Assessments rely on protocols based on fundamental assumptions about the timing of peak aquatic primary production, dominant groups, and spatial distribution. Without a full understanding of aquatic primary producer biomass controls, conclusions about extent and appropriate management controls can be misleading.

Figure III-1. Examples of four major primary producer groups found in tidal flats, shallow, and deepwater habitat types in estuaries: macroalgae on tidal flats (top left) and floating macroalgae in a closed lagoon (top right), seagrass (second panel, left), and *Ruppia spp.* sp., a type of brackish water submerged aquatic vegetation (second panel, right), microphytobenthos (third panel left and right) and phytoplankton (bottom panel left and right). From Sutula (2011).

Estuaries in the SCB are typical of those found in Mediterranean climates such as those found on the Pacific coasts of the United States, Chile, South Africa, and Europe. In general, these estuaries are characterized by a strong seasonality in rainfall and freshwater flow, with high inputs during a winter wet season and low inputs during an extended dry season (Largier *et al.* 1997, Obrador *et al.* 2008, Perez-Ruzafa *et al.* 2011). A common type of estuary found in Mediterranean climates is the bar-built estuary, so called because of the formation of sandbars that build up along the mouth as a consequence of the longshore transport of sand. These sand bars serve to restrict or completely closed the waters behind them from surface water tidal exchange with the ocean (Largier *et al.* 1991, Suzuki *et al.* 1998). Bar-built estuaries are usually shallow (<2 m), with reduced tidal action during time periods when the sand bar restricts tidal exchange, typically during periods of low freshwater input. While Mediterranean bar-built estuaries are generally dominated by benthic aquatic primary producers groups (seagrass, macroalgae, microphytobenthos), few large-scale regional studies have been conducted that can provide comparative data on basic ecological patterns, timing of peak abundances, and factors controlling differences in expression across estuaries, including the effect of estuarine class and degree of tidal exchange across the ocean inlet.

The purpose of this study was two- fold: 1) document the dominant groups and temporal patterns of abundance in aquatic primary producers found in Mediterranean bar-built estuaries of the Southern California Bight, a region found on South West Pacific Coast of the United States, 2) investigate the factors associated with these patterns, including estuarine class and degree of tidal exchange with the coastal ocean, estuarine characteristics such as salinity, sediment particle size and organic matter content, water column nutrient concentrations and ratios. This study took advantage of primary producer abundance and collateral data collected synoptically using standardized methods for 27 sites in 23 estuaries in the Southern California Bight during from November -October 2009 through the Bight Regional Monitoring Program's Estuarine Eutrophication Assessment (URL).

Methods

Study Area

The Southern California Bight (SCB; Figure II-1, Section II) is an open embayment in the coast between Point Conception, California and Cabo Colnett (south of Ensenada), Baja California. The region has a Mediterranean climate, with an average annual rainfall of 10–100 cm (e.g., (Nezlin and Stein 2005)), falling primarily during winter months (December through March), and approximately 20 annual storm events (Ackerman and Weisberg 2003). Approximately 100 watersheds, encompassing fourteen thousand square miles and dominated by urban and agricultural land uses, drain into SCB estuaries and nearshore waters (Ackerman and Schiff 2003). Stormwater runoff to the SCB contributes 95% of the total annual runoff volume (Schiff *et al.* 2000, Ackerman and Weisberg 2003).

SCB estuaries range in size from < 1 to > 5000 hectares. Three estuarine classes or geoforms are represented in this region (Figure III-2): 1) enclosed bays are well flushed with a strong tidal prism and dominated by shallow or deepwater subtidal habitat. The inlet mouth is not restricted and is perennially open to tidal exchange, 2) lagoons and 3) river mouth estuaries have restricted tidal inlets, are dominated by shallow subtidal and intertidal habitat and have a long residence time due to the restricted width of the mouth. Lagoons historically received less freshwater input, though in recent years many receive substantial input from urban runoff; river mouth estuaries are dominated by fluvial forcing. For both lagoons and river mouths, the inlet can be open or closed, perennially (all year round), intermittently (open at least once per year) or ephemeral (opens infrequently, usually every several years or not known recently to open). Among these bar-built estuaries, enclosed bays are the largest class as percent of estuarine habitat, the smaller but more numerous lagoons and river mouth estuaries in California represent 90% by number of the State's estuaries. These estuaries can be more susceptible to degradation by anthropogenic stressors because of lack of flushing that occurs when the sand bar is closed (Valiela *et al.* 1997, Suzuki *et al.* 1998).

In southern California, the conversion of open land into impervious surfaces has included dredging and filling over 75% of bays and estuaries and extensive alterations of coastal streams and rivers (Brownlie and Taylor 1981, Horn and Allen 1985, NRC 1990, Zedler 1996). This "hardening of the coast" changes both the timing and rate of runoff releases to estuaries coastal waters and can affect water quality through addition of sediment, nutrients and other contaminants. It has also dramatically changed the hydrology of estuaries, resulting in the type conversion from one estuarine class to another as well as fragmented sections of estuaries that are hydrologically-isolated behind levees with tidal exchange controlled by tide gates or weirs.

Figure III-2. Examples of three major estuarine geoforms in California: enclosed bay (left), lagoon (center) and river mouth estuary (right).

Bight'08 Eutrophication Assessment Study Design

The SCB Eutrophication Assessment sought to answer the following questions: 1) what the extent and magnitude of eutrophication in SCB estuaries and 2) what the relationship between nutrient loads, estuarine nutrient concentration, and indicators of estuarine eutrophication. The eutrophication assessment was conducted as a probability-based survey in which sites are randomly selected from a comprehensive list of 57 estuaries.

Because this was a regional assessment, data collection emphasized sampling across many estuaries, rather than to better characterize eutrophication spatially within an estuary. Eutrophication is highly spatially variable within an estuary, therefore, an index area was chosen in each system based on 1) proximity to the greatest source of freshwater nutrient loads, 2) zone in which residence time is the longest, and 3) feasibility and safety of access for frequent maintenance. This index area is hereto referred to as a "segment." Because the segment in many cases does not represent the entire estuary, the reporting unit is the estuarine segment. In each of these segments, riverine nutrient loads, primary producer abundance and DO were monitored during November 2008-October 2009.

The sampling design took into account the three subpopulations of interest: 1) estuarine geoform (enclosed bays, lagoons, and river mouth estuaries), 2) tidal regime (perennially, intermittently and ephemerally open to surface water tidal exchange) and 3) presence or absence of anthropogenic muting of tidal regime, defined by 100% containment of the segment by the presence of dikes and levees, with the presence of tide gates or weirs that reduce the amplitude of tidally-induced water level fluctuations in the estuary. These sites were selected as paired sites undiked versus diked within an estuary. While these were not sampled as separate strata in the survey, site selection was weighted to ensure adequate sampling of sub-populations of interest. The sample frame was developed by drawing up a comprehensive list of coastal drainages in southern California coastal watersheds and attributing these estuaries by estuarine class, tidal regime and presence of anthropogenic muting. Some estuaries were excluded from the sample frame. Small creek mouths less than 10 m in width at the mouth, were felt to have insufficient area for assessment purposes. Open embayments, were felt to be driven by physical factors compared to enclosed bays, river mouths, and lagoons and were also eliminated to ensure comparability among sites in the study. Ports and marinas were excluded from the frame because of additional anthropogenic effects on these systems may interfere with expression of eutrophication. Estuaries currently undergoing restoration efforts were also excluded because restoration efforts (dredging, planting, etc.) may mask expression of eutrophication. Table II-1 (Section II) gives the characteristics of the estuaries included in the assessment. A total of 27 segments were selected in 23 estuaries.

Field Methods and Laboratory Methods

Field and laboratory methods are summarized in Section II. Detailed methods and quality assurance measures are provided in detail in Appendix C (QAPP). Data collection methodologies for primary producers as well as collateral data for sediment and water column parameters are briefly summarized below. Sampling of primary producer abundance included macroalgal biomass and percent cover, surface water phytoplankton biomass (chlorophyll *a*), benthic algal biomass (sediment chlorophyll *a*), brackish submerged aquatic vegetation (submerged aquatic vegetation) density and percent cover. Measurement of primary producer communities occurred every other month in all estuaries for a year beginning from November 2008 through October 2009. Continuous water quality monitoring provided chlorophyll *a* fluorescence data, calibrated with surface water measures of chlorophyll *a* made every other month. Seagrass abundance (i.e., *Zostera spp.)* was not assessed because of costs associated with deployment of divers. Instead, existing data on extent from seagrass monitoring program was used to identify segments with appreciable seagrass habitat (>20 acres; Bernstein *et al*. 2011). Table II-2 gives a detailed list of indicators and analytes measured, the data collection method, and the frequency of measurement.

Macroalgae Abundance. Monitoring of macroalgal abundance provided information on when algal blooms occur in each class of estuary, how far they extend spatially, and how long they endure.

Macroalgal abundance was determined by measuring percent cover and algal biomass. Within each index area, three 30 - 50 m transects were laid out in the intertidal area, parallel to the water's edge and along the same elevational contour at approximately three quarters of the distance from the mean lowest low water line to the downslope end of vascular vegetation on the mid-to-upper mudflat. This area has been demonstrated to be representative of macroalgae accumulation in southern California estuaries (Kennison *et al.* 2003). Percent cover was measured at ten randomly chosen points along each transect by placing a 0.5 m^2 quadrat with 49 intercepts on the benthos and recording the presence or absence of each macroalgae species under each intercept. Biomass was collected at 5 of the quadrat locations. Each biomass sample was refrigerated until analysis and processed within 24 hours of collection. In the laboratory, algal samples were cleaned of macroscopic debris, mud and animals, and sorted to genus level. Excess water was shed from each sample, which was then weighed wet, and dried at 60°C to a constant weight, then weighed dry. During data analysis, all macroalgae genus weights were summed for each quadrat to give a total macroalgae wet and dry weight in each quadrat. The average biomass and cover values for all quadrats in a transect were averaged into a single transect value and the three transect values were averaged into a segment value for each segment for each sampling period. Because of lack of confidence in taxonomic expertise among the field groups, a decision was made to lump macroalgal biomass and cover data into broad taxonomic groups (green, red), maintaining wrack separately.

Phytoplankton Biomass. Phytoplankton biomass was estimated from fluorescence measurements collected via *in situ* optical probe (YSI 6600 sonde, chlorophyll fluorescence probe), with bi-monthly discrete chlorophyll *a* water samples taken to calibrate the continuous fluorometry. Discrete suspended chlorophyll *a* pigments were concentrated from 250-500 ml of sample water by filtering at low vacuum through a 45mm diameter Whatman glass fiber filter. Filters were stored in a petri-dish covered in aluminum foil and frozen until analysis. Photosynthetic pigments were extracted from filters in 90% acetone solution and allowed to steep overnight, to ensure complete extraction of chlorophyll *a* (EPA 445). Fluorescence was measured before and after acidification with 0.1 M HCl to determine the phaeophytin-corrected chlorophyll *a*. Concentrations were calculated relative to a laboratory standard. *In situ* chlorophyll fluorescence was measured every 15 minutes using an optical probe mounted to a YSI 6600 V2 data sonde. Probes were maintained according to factory specifications and were routinely calibrated. Fluorescence measurements were calibrated to chlorophyll *a* concentrations using least-squares regression generated from daily averaged data probe measurements and discrete concentration data collected on that same day.

Microphytobenthos. Benthic chlorophyll *a* (microphytobenthos) was determined on sediment composite samples collected at the beginning and end of each macroalgae transect. Composites were comprised of ten sediment plugs (3 cm in diameter, 1 cm deep) collected at each end of the transect (20 total). The samples were collected downslope of the end of each transect in approximately 30 cm of water depth. Plugs were homogenized in sample bags and refrigerated until analysis. A subsample from each bag was collected in a 15 ml centrifuge tube and frozen for benthic chlorophyll *a* analysis. Benthic chlorophyll *a* samples were analyzed for chlorophyll *a* and phaeopigments as described above for phytoplankton.

***Ruppia spp.* Abundance.** Monitoring of *Ruppia spp.* (brackish water submerged aquatic vegetation) abundance provided information on when and where stands occur in each class of estuary, how far they extend spatially, and how long they endure. *Ruppia spp.* abundance was determined by measuring biomass. Within the subset of estuaries where *Ruppia spp.* was observed in each index area, three transects were laid out perpendicular to the shore-line bisecting the channel. Biomass was collected using sampling "tongs" (Rodusky *et al.* 2005), comprehensively collecting all biomass in a defined area, at 3 to 5 across the channel (at the thalweg, and one or two evenly spaced locations between the thalweg and the water's edge). Each biomass sample was refrigerated until analysis and processed within 24 hours of collection. In the laboratory, biomass samples were cleaned of macroscopic debris, mud and animals, and sorted to genus level. Excess water was shed from each sample, which was then weighed wet, and dried at 60°C to a constant weight, then weighed dry.

Water Column Physiochemistry. Water column physiochemistry was measured continuously using a YSI 6600 data sonde. Each sonde was outfitted with a conductivity/ temperature sensor, ROX optical dissolved oxygen probe, extended deployment pH probe, chlorophyll optical sensor, and a turbidity optical sensor. All sensors were treated with anti-fouling tape and calibrated at a minimum of once monthly. Sondes were deployed at one location in each segment, in bottom water (approximately 30 cm from the sediment surface). Measurements were collected every 15 minutes throughout the deployment period. Dissolved oxygen concentrations were calculated from percent saturation, temperature and salinity data. An hourly running average was applied to the data set to smooth high frequency noise.

Ambient Nutrients. A single grab sample was collected and subsampled for ambient nutrients (nitrate, nitrite, ammonium, soluble reactive phosphate, total nitrogen, total phosphorus, total dissolved nitrogen and total dissolved phosphorus). A 1 L amber bottle which was triple rinsed with sample water before filling completely. The sample bottle was open and closed under water to avoid contamination with surface films. The bottle was subsampled for a suite of analytes using a clean, 60 ml syringe triple rinsed with sample water. Dissolved inorganic and total nutrients were filtered through a 0.45 μm mixed cellulose ester (MCE) filter rinsed with 20 ml of sample water (discarded) before collection into triple rinsed 30 ml HDPE sample bottles. One subsample was assayed by flow injection analysis for dissolved inorganic nutrients using a Lachat Instruments QuikChem 8000 autoanalyzer for the analysis of NH_4, NO_3, NO_2, and SRP. Subsamples for TDN, TDP, TN and TP were assayed via two-step process: first water samples undergo a persulfate digest to convert all N into NO_3 and all P into orthophosphate; then the resulting digests were analyzed by automated colorimetry (Alpkem or Technicon) for nitrate-N and orthophosphate-P (Koroleff 1985).

Sediment Nutrients and Grain Size. Sediment characteristics and benthic chlorophyll *a* (benthic microalgae) were determined on sediment composite samples collected at the beginning and end of each macroalgae transect. Composites were comprised of ten sediment plugs (3 cm in diameter, 1 cm deep) collected at each end of the transect (20 total). The samples were collected downslope of the end of each transect in approximately 30 cm of water depth. Plugs were homogenized in sample bags and refrigerated until analysis. A subsample from each bag was collected in a 15 ml centrifuge tube and frozen for benthic chlorophyll *a* analysis. The remaining sediment was transferred to an aluminum dish, weighed wet and dried at 60°C to a constant weight and weighed dry. A subsample of dried sediment was ground with a mortar and pestle for analysis of percent total phosphorus (%TP), percent total nitrogen (%TN) and % organic carbon (%OC). Samples for %OC were acidified to remove carbonates; %OC and %TN were measured by high temperature combustion on a Control Equipment Corp CEC 440HA elemental analyzer at the Marine Science Institute, Santa Barbara. Sediment %TP were prepared using an acid persulfate digest to convert all P to orthophosphate that was then analyzed by automated colorimetry (Technicon) at the University of Georgia Analytical Chemistry Laboratory. The remainder of the sediment was reweighed dry, wet sieved through a 65µm sieve, dried at 60°C to a constant weight, and weighed dry to determine grain size. Percent fines were calculated using the difference between the total weight and the weight of the sieved portion. Benthic chlorophyll *a* samples were analyzed for chlorophyll *a* and phaeopigments as described above for phytoplankton.

Data Analysis

Sources of Collateral Data on Factors

We utilized three sources for collateral data to analyze the effect of estuarine "factors" on primary producer biomass: 1) summarized estuarine area associated with specific estuarine habitat types, generated from wetland and seagrass maps (www.socalwetlands.org; Bernstein *et al*. 2011), 2) merged bathymetry/topography data generated as a part of a study of estuarine classification (Sutula *et al*. unpublished data). These data sets were used to describe for each estuarine segment the following parameters:

Analysis of Tidal Inlet Forcing Using Discrete Fourier Transform (DFT). The contribution of semi-diurnal and diurnal tidal forcing regulating primary producer variability was investigated using DFT. For this, each water-level continuous data sonde time series was interpolated on regular 15-min time intervals and DFT power spectra were calculated for all estuaries for the entire period of observations using standard MATLAB routines. All spectra demonstrated two dominating peaks of variability in water surface elevation (WSE) and dissolved oxygen: diurnal (period ~1 day) and semi-diurnal (period ~0.5 day) . The magnitude of each peak was extracted from the DFT power spectrum (diurnal peak at 0.9–1.1 days/cycle and tidal peak at 0.4–0.6 days/cycle). Distribution of diurnal and semi-diurnal power spectra intensity among SCB estuaries was used as both a continuous variable as well as a means to categorize the estuaries with respect to tidal inlet status.

Statistical Analysis. Statistical relationships between primary producer communities and co-factors were assessed over a variety of timescales: annual (water year: Nov 2008- Oct 2009), wet season (Nov 2008- Apr 2009), dry season (May 2009-Oct 2009), as well as on a sampling period by period basis. Peak season is defined as the average biomass/cover of segment values for the two consecutive periods of highest biomass/cover. The "maximum" period is defined as the highest single period of biomass/cover. Means, standard errors, population maximum, and population minimums were all calculated using MS Excel.

Exploratory Analysis. To investigate relationships between primary producer biomass and cofactors during each sampling period we used non-parametric statistics (Wilcoxon signed rank test and

Spearman's rho) on untransformed data. To test for relationships among factors we used least-squares regressions on log-transformed data. Statistical tests were conducted on JMP 9.0 (SAS Institute Inc.).

Estuarine Geoform and Inlet Status Classification. We used a conceptual approach modeled after the Coastal Marine Ecological Classification Standard (CMECS; (Madden *et al.* 2005)) to classify the segments according to dominant geoform (enclosed bays, lagoons, and river mouth estuaries). We also classified estuaries according to the status of their respective ocean inlets (perennially open, intermittent/ephemeral, and anthropogenically muted) (Table II-1). We used a two way, repeated measures analysis of variance (ANOVA factors: sample period and class; sample period and inlet status) to determine if primary producer response was related to either estuarine geoform or inlet status (sample size was insufficient for a three way ANOVA). The influence of the presence of dikes or weirs on primary producer expression was investigated through analysis of differences between paired sites in the same estuarine complex. These paired sites were located in San Diego Bay (SDM and SDF), Bolsa Chica (BCM and BCF), Anaheim Bay/Seal Beach (SBM and SBF), and Mugu Lagoon (MLM and MLF). Statistical tests were conducted on JMP 9.0 (SAS Institute Inc.).

Biomass Comparison. In order to compare biomass among the primary producer groups, biomass was converted to carbon content. Macroalgae and *Ruppia spp.* dry biomass was converted to carbon content assuming biomass was 22% C by dry weight (Wetzel *et al.* 1981, Lapointe *et al.* 1992). Phytoplankton and benthic microalgae were converted to carbon content by assuming a Chla:C ratio of 30 (Geider *et al.* 1997, Geider and LaRoche 2002).

Results

Temporal and Spatial Patterns in Aquatic Primary Producer Biomass: Comparison across SCB Estuaries

Macroalgae, phytoplankton, and benthic microalgae had detectable biomass in all segments surveyed. Though biomass varied across segments and with season, all types of aquatic primary producers were found in the SCB in all sampling periods and over a range of salinities (Table III-1, Figure III-3). Brackish submerged aquatic vegetation (*Ruppia spp.)* was present in 30% of the systems. In general, macroalgae dominated aquatic primary producer biomass during sampling periods (Figure III-1), with mean annual biomass of 47 ± 50 g dw m^{-2} across segments, and segment means ranging from 0.4 to 154 g dw m^{-2}.

Table III-1. Mean, standard error, coefficient of variation, minimum value and maximum value for macroalgae dry biomass, macroalgae cover, microphytobenthos biomass, phytoplankton biomass, and *Ruppia spp.* dry biomass during each sampling period across the southern California Bight

Period	Macroalgae Biomass (g dw m^{-2})				Macroalgae Cover (%)				Microphytobenthos (Benthic Chl *a*) (mg m^{-2})				Phytoplankton (Suspended Chl *a*) (mg m^{-3})				*Ruppia spp.* (g dw m^{-3})			
	Mean ±SE	CV	Min	Max	Mean ±SE	CV	Min	Max	Mean ±SE	CV	Min	Max	Mean ±SE	CV	Min	Max	Mean ± SE	CV	Min	Max
Nov-08	52.5 ±71.9	1.4	0.0	248.1	34.2 ±28.3	0.8	0.0	92.9	39.7 ±158.0	4.0	0.5	830	7.05 ±10.26	1.5	0.75	19.89	40.3 ±68.0	1.7	0.0	190.3
Jan-09	20.0 ±36.2	1.9	0.0	137.8	19.3 ±22.9	1.2	0.0	82.3	652.5 ±752.9	1.2	12.5	2901	7.80 ±12.36	1.6	0.35	42.88	7.3 ±12.3	1.7	0.0	30.0
Mar-09	29.9 ±44.9	1.5	0.0	211.1	29.2 ±28.4	1.0	0.0	94.4	1482.5 ±907.9	0.6	537.2	3527	10.26 ±19.52	1.9	0.70	91.37	9.0 ±24.0	2.7	0.0	68.3
May-09	61.1 ±80.2	1.3	0.0	285.2	31.7 ±27.9	0.9	0.0	91.6	938.1 ±759.9	0.8	176.1	3296	8.15 ±9.26	1.1	0.59	33.03	16.8 ±22.2	1.3	0.0	66.0
Jul-09	39.3 ±64.1	1.7	0.0	286.7	28.2 ±27.2	1.0	0.0	94.4	1246.3 ±1519.7	1.2	216.9	7557	7.88 ±9.60	1.2	0.56	32.43	408.0 ±822.4	2.0	0.0	2359
Sep-09	61.4 ±135.9	2.3	0.0	184.4	29.9 ±23.8	0.8	0.0	86.5	868.0 ±647.9	0.8	8.5	2630	6.07 ±9.20	1.5	0.45	39.63	757.7 ±863.5	1.1	0.0	2413

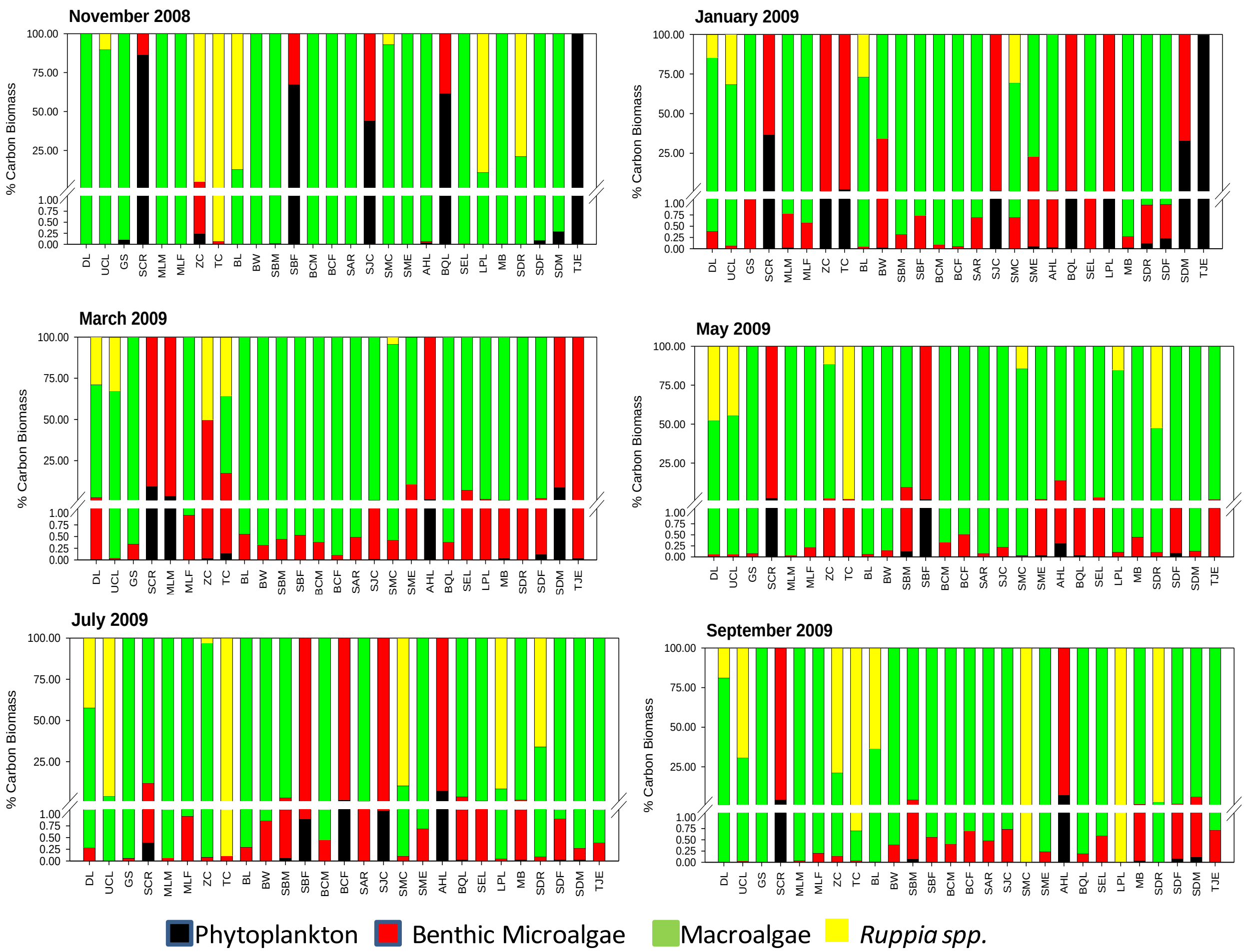

Figure III-3. Representation of relative contribution of each group to standing stock of aquatic primary producers carbon biomass, normalized to a g C m^{-2}. Each panel represents a sampling period, from November 2008 through September 2009, with the X axis representing estuarine segments, from north to south. Y axis is % of biomass as carbon.

Wrack, which is primary producer biomass not generated within the estuary but rafted in from upstream or the coastal zone and consisting mainly of marine kelp species (e.g., *Macrosystis spp*), was a dominant component of the macroalgae biomass found in some estuaries (e.g., DL, SBM, SEL), but was excluded from biomass in this study. Macroalgal biomass was for the most part completely dominated by green algae (*Ulva spp.*). Red macroalgae (e.g., *Ceremium spp. and Gracilaria spp.*) was occasionally found in a few segments (BCM, MB, SDM, UCL), typically in estuaries with salinities > 34 ppt (Figure III-4).

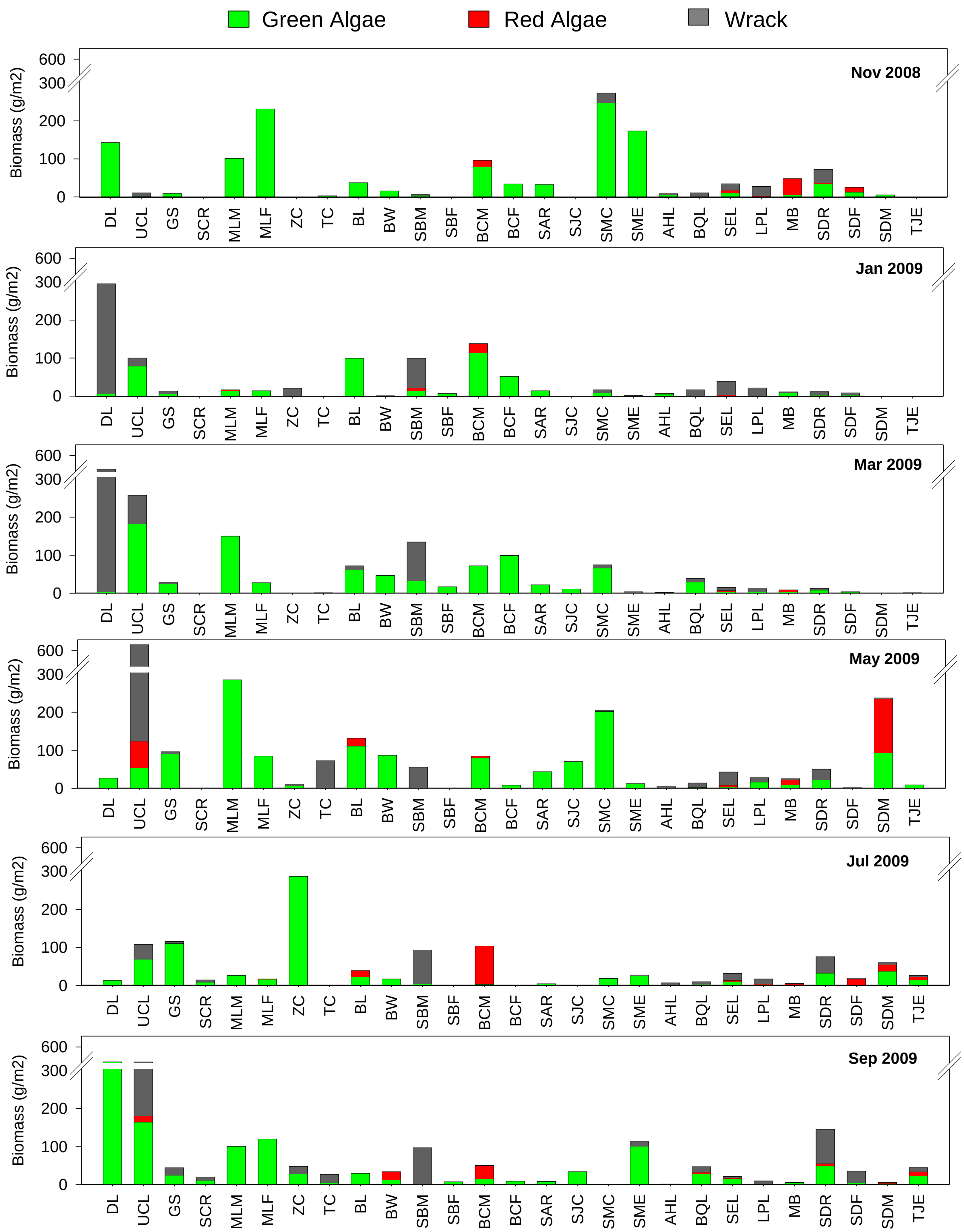

Figure III-4. Biomass of red and green macroalgae relative to wrack in intertidal transect. Each panel represents a sampling period, from November 2008 through September 2009, with the X axis representing estuarine segments, from north to south.

Across SCB segments, annual mean macroalgal cover was 30 ± 22 %, ranging from 0 to 82%. When present, brackish submerged aquatic vegetation was a significant fraction of the carbon biomass of the segment at its peak productivity (Figure III-4), with mean annual biomass of 73 ± 149 g dw m^{-3} and ranges from 0 - 467 g dw m^{-3}. Phytoplankton was generally low in most systems (annual mean of 10 ± 18 µg Chl *a* L^{-1} across segments) or, at most, co-dominant with macroalgae. The exception to this was Santa Clara River estuary, where phytoplankton was dominant for most of the year, or co-dominant with microphytobenthos. Microphytobenthos was seasonally important in many estuaries, with a mean of 890 ± 549 µg Chl *a* m^{-2} across segments and a range of 247 - 2030 µg Chl *a* m^{-2}.

Temporal Variability. Aquatic primary producer biomass was highly variable, across estuaries and among seasons. All primary producer groups exhibited distinct seasonal peaks in biomass (Figures III-4 and III-5). For all segments, macroalgae, microphytobenthos, and *Ruppia spp.* (where present) were found to have at least one or more sampling periods having significantly higher biomass than the remaining periods, though these seasonal peaks were not consistent among all segments (Figure III-3). Seasonality was greatest for microphytobenthos and brackish submerged aquatic vegetation. Microphytobenthos peaked in the March 2009 in 75% of segments, while *Ruppia spp.* biomass peaked in summer through late fall (July - November), with 65% of segments peaking in September 2009. With phytoplankton, peak chlorophyll *a* typically occurred in the spring, with 70% of segments showing peak biomass in either March or May 2009. Across segments, seasonality was somewhat less distinct for macroalgae, with 30% peaking in May 2009, and 20% in September and March 2009.

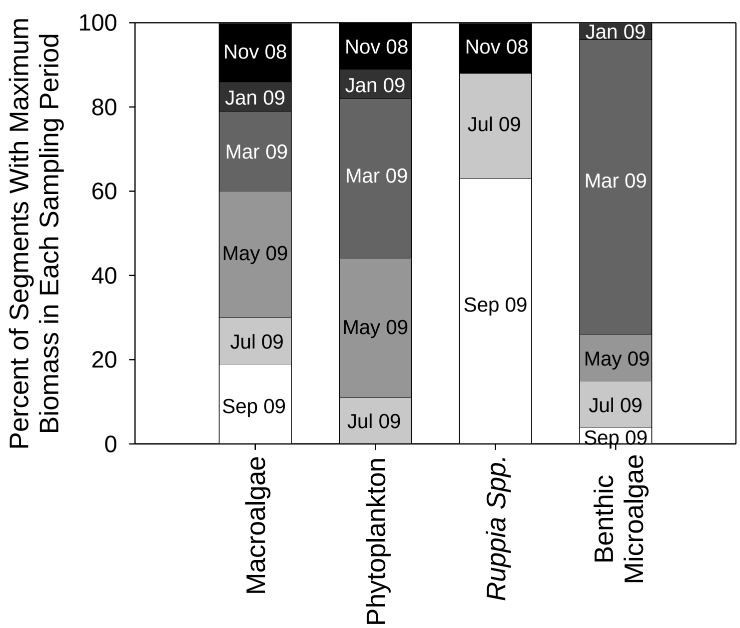

Figure III-5. Relative percentage of segments with maximum biomass in each of the six sampling periods. Period of maximum biomass is variable by segment and primary producer group.

Across all SCB segments, macroalgae biomass was significantly higher in the spring and summer compared to the winter ($p < 0.05$, Wilcoxon paired test), with January 2009 significantly lower than May, July, and September 2009; Table III-1). Macroalgae cover was significantly higher in November 2008, September 2009, and May 2009 compared to January 2009. However, not all systems had peak biomass and cover during the summer and fall. Several segments had their peak biomass and/or cover during the winter (January and March) sampling periods (Figure III-5). Generally, high biomass was positively correlated with high percent cover ($R^2 = 0.51$, $p < 0.001$; Figure III-6). However, there were periods where peak biomass and peak cover did not coincide. Thirty-three percent of segment sites had a period of peak biomass that was decoupled from its period of peak cover. In these cases, the period of peak cover preceded the period of peak biomass by 1 or 2 sampling periods (2-4 months). Furthermore, periods of high cover (>25%) and low biomass (<50 g dw m^{-2}) were common (23% of all sampling events), though periods of high biomass (> 70 g dw m^{-2}) and low cover (< 15%) were rare (1% of all events). Of the systems that had high biomass (52%), 66% of these segments had more than one sampling period of high biomass (at least 8 weeks) suggesting a chronic condition (Appendix D).

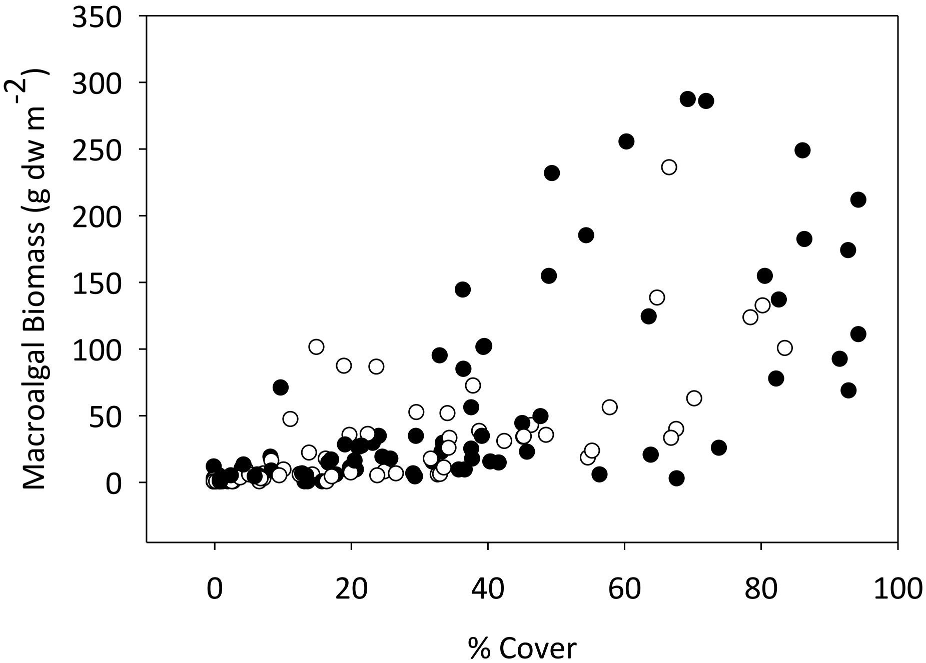

Figure III-6. Relationship of mean macroalgal biomass versus mean percent cover for all segment sites for all index periods. Black circles represents segments in estuaries "closed" to surface water tidal exchange; open circles represent segments in estuaries with "open " to surface water tidal exchange.

Microphytobenthos biomass across all SCB segments was significantly different by season (Wilcoxon signed rank test, $p < 0.05$), with the lowest biomass in November 2008 (mean of 39.7 ± 158 mg Chl *a* m^{-2} across segments;) and greatest in March 2009 (1483 ± 908 mg Chl *a* m^{-2} ; Table III-1). Most segments (70%) had peak benthic chlorophyll *a* concentrations in March 2009, which was typically before most of the segment sites began experiencing their highest macroalgae and/or *Ruppia spp.* biomass (Figure III-5, Appendix D).

Bight-wide, phytoplankton biomass (measured as suspended chlorophyll *a*) had no single period that was significantly different from other sampling events (Wilcoxon signed rank test, $p < 0.05$). Overall, most segments peaked in the late winter/early spring (70% of segments) and no segments had peak biomass in September (Figure III-5). Twenty-eight percent of SCB segments had Chl *a* concentrations greater than 10 μg L^{-1} for more than 10 % of the deployment (>4 weeks); 21% of all SCB segments had chlorophyll *a* concentrations greater than 10 μg Chl *a* L^{-1} for longer than 8 weeks (Figure III-7; Appendix D).

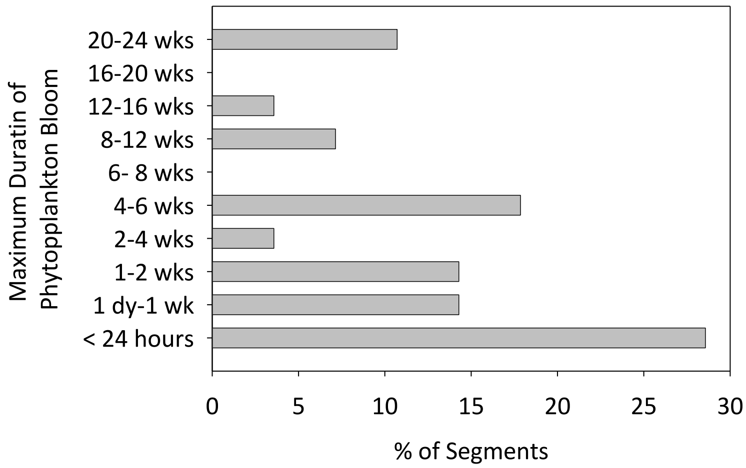

Figure III-7. Histogram of duration of elevated phytoplankton biomass (>10 μg chl a L^{-1}) by frequency of event.

Brackish-water submerged aquatic vegetation (*Ruppia spp.*) was only present in a subset of systems (30%). Bight-wide, there were significant differences in *Ruppia spp.* biomass by season (Wilcoxon signed rank test, $p < 0.05$). Peak biomass occurred latest in the year compared with other primary producer groups, with lowest values in early spring, increasing to a peak in early fall (September), Figure III-5, Appendix D). Half of the sites in which *Ruppia spp.* was present experienced a period of ocean inlet closure, and biomass increased from the time of closure to the last sampling period in September when the inlet was still closed (inlets typically open during storm events in the winter). In these systems, *Ruppia spp.* biomass was not present during the time when the system was open and well flushed. Three of the systems with *Ruppia spp.* were open to tidal exchange throughout the year (one of these systems had a muted tidal regime due to the presence of a tide-gate). In all of these systems, there was no *Ruppia spp.* biomass during the winter months when freshwater flow was highest. Two of the systems with *Ruppia spp.* were continuously closed to tidal exchange throughout the study and these systems had observable biomass during every sampling period, though biomass was greatest in the late summer and fall.

Spatial Variability. Generally, there were no north-south trends evident in aquatic primary producer biomass. Primary producers measured in transects (macroalgae, microphytobenthos, and brackish submerged aquatic vegetation) exhibited spatial variability, with this patchiness generally increasing with the average biomass by transect. For example, with macroalgae, variation in biomass increased with increasing biomass (Figure III-8, top panel). Spatial variability in phytoplankton could not be assessed because sampling only occurred at one location in the segment.

For macroalgae, there was a distinct difference in behavior of biomass versus percent cover. Among the 27 segments, some had consistently high biomass at all transects and other segments where one or two transects had high biomass and the remaining transects had low biomass (explaining the scatter in the coefficient of variability; Figure III-8, bottom panel). This does not appear to be the case for macroalgae percent cover; cover typically increases consistently throughout the segment (decreasing trend in coefficient of variability). Variability in biomass was greatest in September, whereas variability in cover was fairly consistent throughout the year.

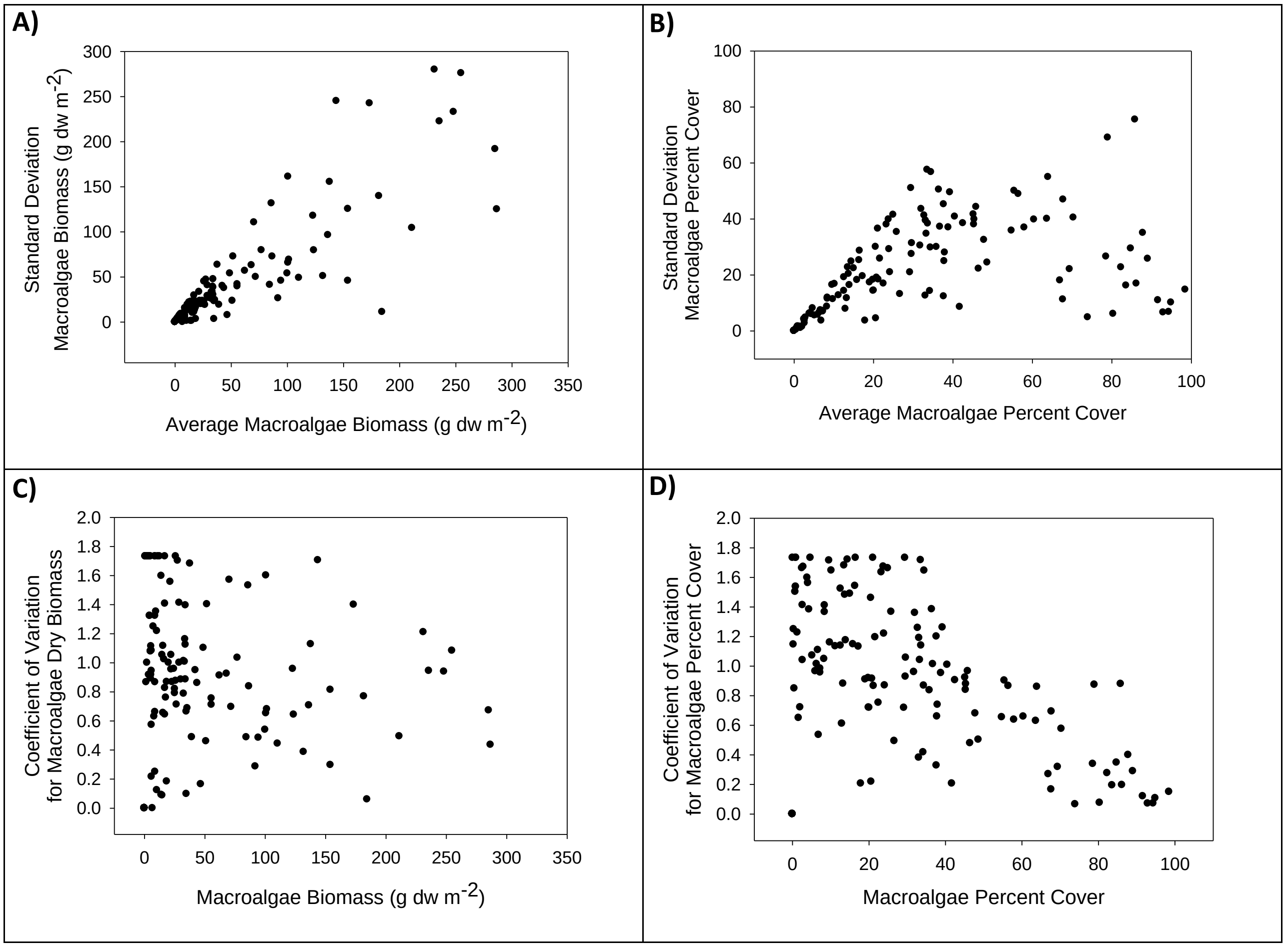

Figure III-8. Average macroalgae biomass as a function of the standard deviation (A) and coefficient of variability (C) and macroalgae percent cover as a function of the standard deviation (B) and coefficient of variability (D) for each segment site during all sampling periods.

Factors Associated with Trends in Aquatic Primary Producer Biomass

Estuarine Geoform, Inlet Status, and Habitat Type. No significant effects were found by estuarine class (enclosed bay, lagoon, river mouth) for macroalgae biomass, phytoplankton biomass, or *Ruppia spp.* biomass (Table III-2). Estuarine class had a strongly significant effect only on microphytobenthos (as sediment chlorophyll *a*), where segments of river mouth estuaries approximately twice the microphytobenthos biomass enclosed bays and lagoons (p-value = 0.021).

Table III-2. Summary of significant two factor ANOVA of the effects of sampling period, inlet status, and class on microphytobenthos. Tests conducted on other aquatic primary producer biomass (macroalgae, phytoplankton and *Ruppia spp.*) were not significant.

Parameter	Source	df	value	exact F	Probability
Benthic CHLa	**inlet**	**2**	**0.358**	**4.297**	**0.025**
	time	**5**	**3.878**	**15.511**	**<0.0001**
	inlet x time	10	0.690	1.000	0.461
	class	**2**	**0.381**	**4.569**	**0.021**
	time	**5**	**3.529**	**14.114**	**<0.0001**
	class x time	10	0.512	1.592	0.145

When inlet status was described as a categorical variable (open, closed, diked), no significant effects were found for macroalgal biomass and cover, phytoplankton biomass, nor *Ruppia spp.* biomass (Table III-2). However, inlet status can also be described as a continuous variable by using the strength of frequencies in semi-diurnal or diurnal water surface elevation (WSE) variations driven by tidal forcing. This indicator is derived from power spectra analysis of fourier-transformed WSE (a.k.a. spectral power intensity or SPI of WSE). Diurnal SPI increases with increasing exchange with ocean at the tidal inlet. We found that phytoplankton biomass, macroalgal biomass, and microphytobenthos biomass were all significantly, negatively correlated with diurnal SPI of WSE. This relationship was strongest for annually averaged macroalgal biomass (p-value = 0.0003, R^2 = 0.43), phytoplankton biomass and microphytobenthos biomass were less strong, but still significant (p-value = 0.0275, R^2 = 0.18; p-value = 0.0174, R^2 = 0.20 for phytoplankton and microphytobenthos respectively). For *Ruppia spp.* biomass, the relationship with SPI could not be evaluated, as 6 of 8 segments with *Ruppia spp.* biomass had an SPI of approximately zero, indicating no water level variation (closed inlet).

Dominant habitat type (open intertidal, open subtidal unvegetated, open seagrass, and closed subtidal) was a better predictor of primary producer biomass than class. We found a statistically significant difference in annual macroalgal biomass by dominant habitat type in the segment (p-value=0.183, R^2 = 0.037), where unvegetated open subtidal and closed brackish submerged aquatic vegetation dominated habitat had 6 times the biomass (47-49 g dw m^{-2}) as seagrass dominated habitats (9 g dw m^{-2}), while intertidally dominated habitats had a wide range of biomass (annual mean of 30 g dw m^{-2}) that was not significantly different from the other habitat types. A weakly significant difference was found for microphytobenthos (p-value = 0.05, R^2 = 0.27), where segments found in open, unvegetated subtidal habitat was approximately twice the biomass as found in segments in seagrass-dominated estuaries. No significant difference was found for phytoplankton (p-value = 0.57). As with diurnal SPI, 6 of 8 segments with *Ruppia spp.* were segments with closed, subtidally dominated estuaries.

We also explored the effect of hydromodification from the presence of dikes and tide gates on aquatic primary producer biomass by looking at paired segments (tidal restriction by dikes, tide gates or weirs and unrestricted tidal flushing) within four estuaries. The paired sites showed no significant differences in macroalgal biomass, but significant differences were detected in phytoplankton biomass such that diked sites typically had higher biomass compared to fully tidal sites (p-value = 0.0306) for three out of the four paired sites. No significant difference was found for microphytobenthos biomass (p-value = 0.56).

Associations among Aquatic Primary Producers Groups. We tested the effect of macroalgal biomass and cover and phytoplankton biomass on microphytobenthos biomass among 27 segments. Using annually-averaged data, no significant effect was observed among these variables (p-value> 0.05).

Salinity and Temperature. Spearman's correlation was used to investigate the relationships between temperature, salinity, and aquatic primary producer biomass. Macroalgae and *Ruppia spp.* were not significantly correlated with temperature or salinity (p-value >0.05). Benthic microalgae was not significantly correlated with temperature but was negatively correlated with salinity during all sampling period, and significantly so during November 2008 and July 2009. Phytoplankton had a significant negative correlation with temperature during March and May (p <0.03) and a significant positive correlation with salinity from January to September (p <0.030).

Surface Water Nutrient Concentrations and Ratios. With the exception of microphytobenthos, all aquatic primary producer groups showed significant relationships with water column nutrient concentrations, depending on temporal scale in which the data were averaged (annual average, wet season (November-April) or dry season (May-October; Table III-3).

Macroalgal biomass was significantly, positively correlated with TN, TDN and TP; the least-squares fit was highest with annually-averaged biomass and nutrient concentration data (R^2 from 0.15 for TP to 0.33 for TN; Table III-3).

Phytoplankton biomass was significantly, positively correlated with TN, TDN and DIN; the least-squares fit was highest for dry season-averaged biomass and nutrient concentration data (R^2 from 0.16 for TP to 0.48 for TN; Table III-3), but also high for wet season averaged data as well.

Ruppia spp. biomass showed a significant positive correlation with annually-averaged and dry season averaged TP and TDP concentrations. The least-squares fit was highest for annually averaged data (R^2 from 0.22 for TDP to 0.24 for TP; Table III-3).

Overall, ratios of TN:TP, TDN: TDP and DIN:DIP in estuaries with an open inlet (both open and unrestricted and open but restricted through presence of tide gates or dikes) indicate that the majority of primary production was nitrogen limited (TN:TP = 10 ± 19, TDN:TDP = 11 ± 24, and DIN:DIP = 13 ± 27; Figure III-9). The exception to this were the segments located in closed estuaries, which were typically P-limited regardless of whether total, total dissolved or dissolved inorganic ratios were used (TN:TP = 402 ± 1309, TDN:TDP = 57 ± 130, and DIN:DIP = 259 ± 719; Figure III-10).

Table III-3. Results of least-squares regression of aquatic primary producer biomass and nutrient concentrations, averaged on different time scales ((annual average, wet season (November-April) or dry season (May-October).

Aquatic Primary Producer	Nutrient Species	Annual Average		Wet Season Average		Dry Season Average	
		R^2	p-value	R^2	p-value	R^2	p-value
Macroalgae	TN	**0.3338**	**0.0020**	0.0098	0.6304	**0.2714**	**0.0064**
	TDN	**0.2953**	**0.0041**	0.0091	0.6423	**0.2891**	**0.0046**
	DIN	0.0832	0.1525	0.0283	0.4112	0.0133	0.5755
	TP	**0.1576**	**0.0447**	0.0580	0.2359	0.1249	0.0766
	TDP	0.0824	0.1550	0.0559	0.2450	0.0507	0.2687
	PO_4	0.0716	0.1864	0.0043	0.7508	0.1295	0.0709
Phytoplankton	TN	**0.4573**	**0.0001**	**0.3629**	**0.0009**	**0.4816**	**<0.0001**
	TDN	**0.3785**	**0.0006**	**0.3387**	**0.0014**	**0.3569**	**0.0010**
	DIN	**0.2050**	**0.0177**	**0.1760**	**0.0294**	**0.1668**	**0.0344**
	TP	0.1023	0.1038	0.0565	0.2325	0.0621	0.2102
	TDP	0.0447	0.2898	0.0068	0.6817	0.0230	0.4503
	PO_4	0.0643	0.2019	0.0001	0.9565	0.0932	0.1214
Microphytobenthos	TN	0.0615	0.2124	0.0226	0.4546	0.0706	0.1803
	TDN	0.0546	0.2407	0.0181	0.5029	0.0485	0.2694
	DIN	0.0530	0.2478	0.0031	0.7815	0.0912	0.1257
	TP	0.0200	0.4817	0.0002	0.9384	0.0248	0.4326
	TDP	0.0316	0.3752	0.0020	0.8257	0.0353	0.3482
	PO_4	0.0038	0.7591	0.0088	0.6416	0.0010	0.8766
Ruppia spp.	TN	0.0007	0.8922	0.0001	0.9672	0.0010	0.8778
	TDN	0.0002	0.9438	0.0001	0.9658	0.0027	0.7983
	DIN	0.0236	0.4441	0.0167	0.5211	0.0599	0.2184
	TP	**0.2436**	**0.0089**	0.0001	0.9655	**0.1768**	**0.0290**
	TDP	**0.2223**	**0.0130**	0.0003	0.9363	**0.1559**	**0.0415**
	PO_4	0.0435	0.2965	0.0185	0.4982	0.0219	0.4616

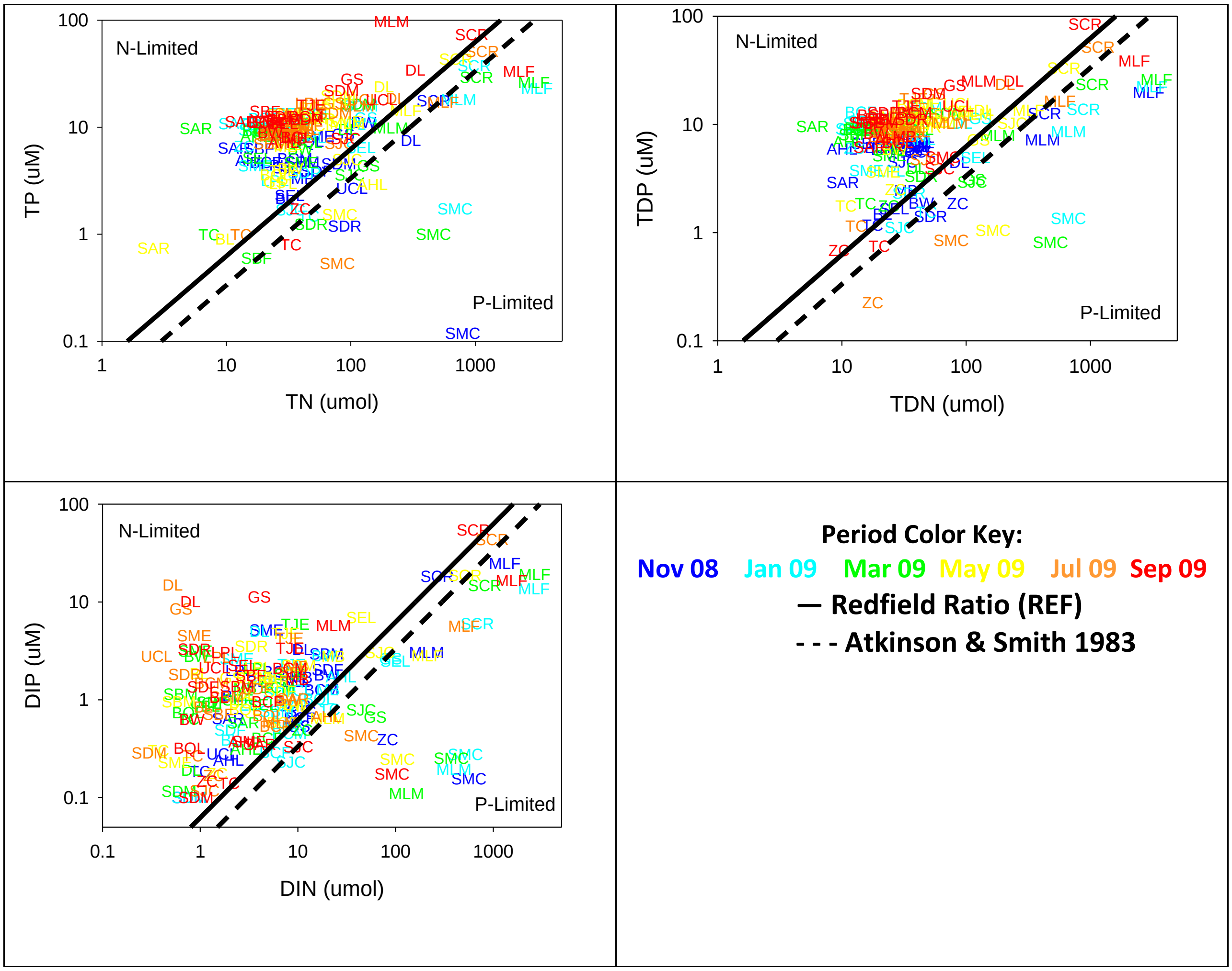

Figure III-9. Nutrient ratios for each segment site for each sampling period compared to the Redfield ratio for phytoplankton (C:N:P = 106:16:1) and the Atkinson and Smith ratio for macroalgae and plants (C:N:P = 550:30:1).

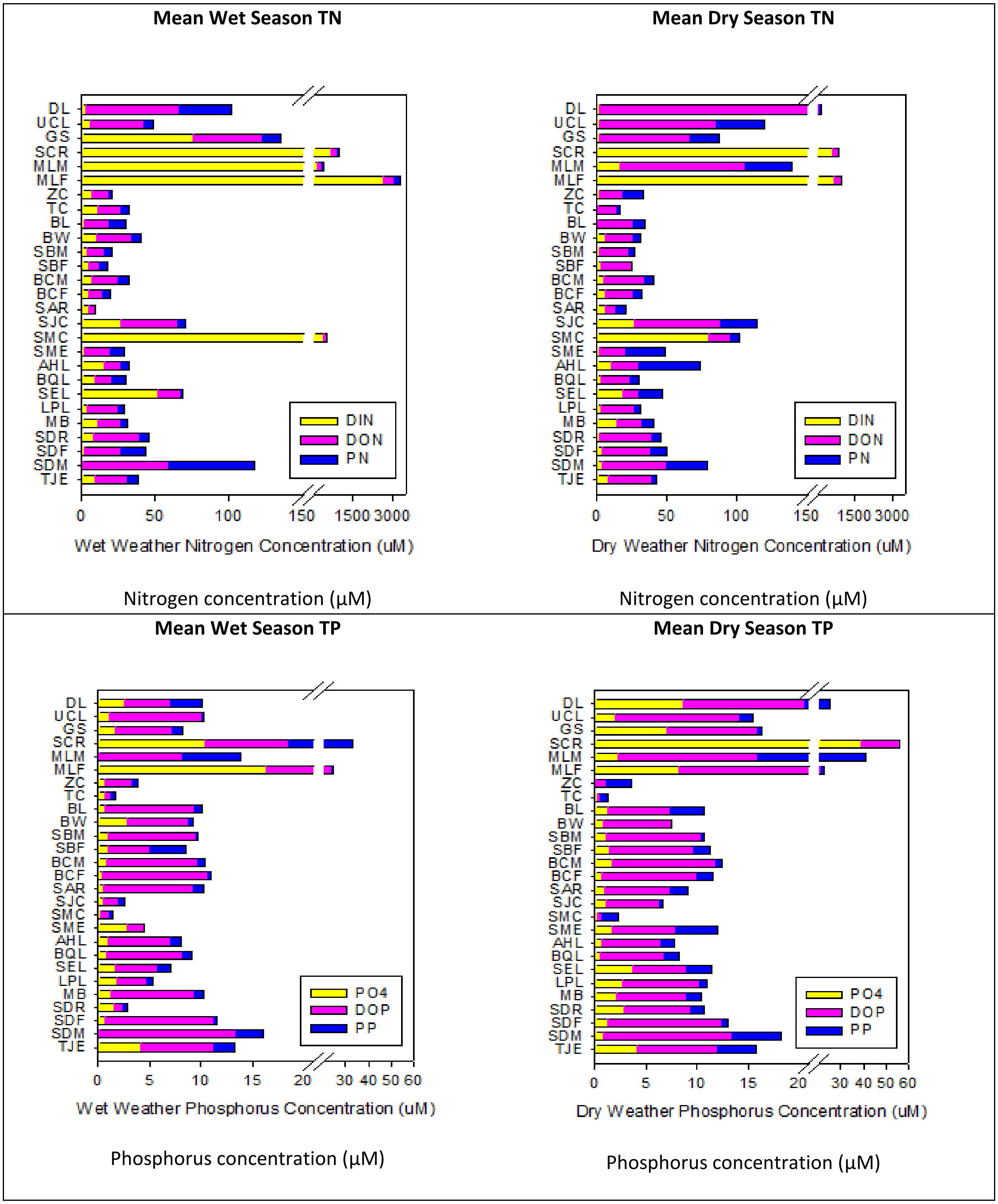

Figure III-10. Relative percent of segment water column TN and TP concentrations as dissolved inorganic species versus organic form by segment site.

Sediment Organic Carbon, Nitrogen and Phosphorus Content. Annual mean macroalgae biomass showed a strong positive correlation with mean %OC (p-value = 0.005, R^2 = 0.27), %TN (p-value = 0.003, R^2 = 0.30; Figure III-9), and %TP (p-value = 0.047, R^2-0.14). While microphytobenthos had no significant relationship with %OC, %TN and %TP, it had a significant negative relationship with increasing percent fines (p-value = 0.0025, R^2-0.31; Figure III-11) and increasing C:N ratio (p-value = 0.0013, R^2 = 0.31). Phytoplankton and *Ruppia spp.* biomass and had no significant relationships with sediment characteristics (p-value>0.05).

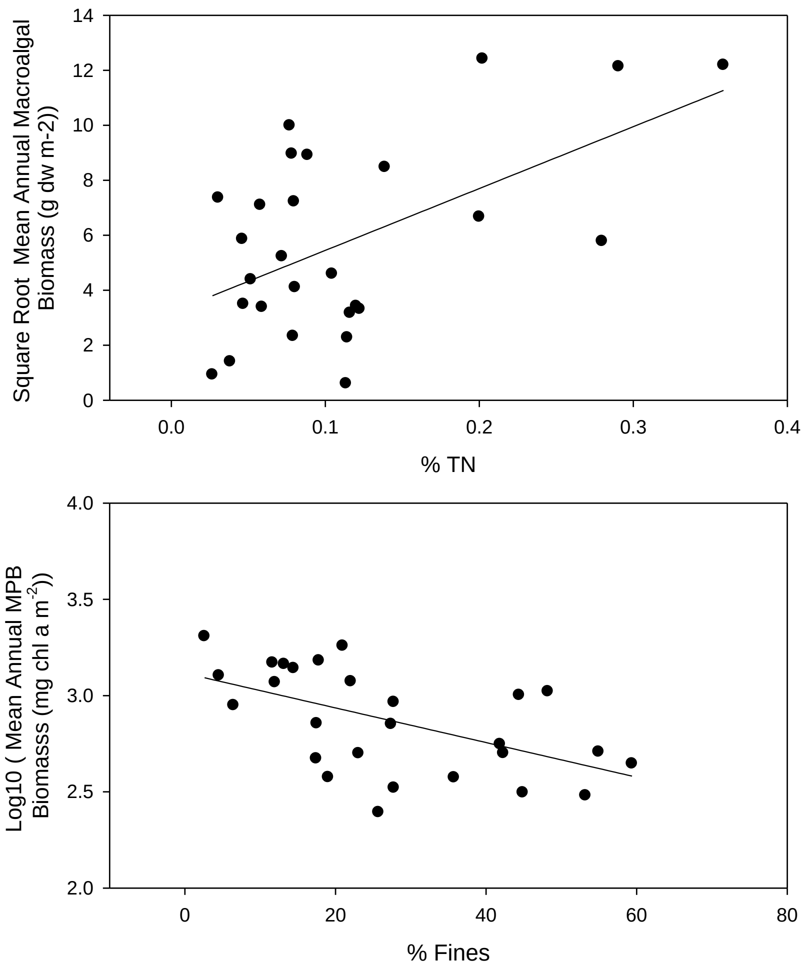

Figure III-11. Least-squares regressions for relationship between sediment %TN and square root of annual mean macroalgal biomass (top panel) and sediment % fines and log_{10} of mean annual microphytobenthos biomass (bottom panel).

Discussion

SCB estuaries represent a wide range in environmental gradients in depth, light availability, residence time, freshwater flow, nutrient and organic matter loading, and sediment characteristics. These factors contribute to the observed temporal patterns in relative abundance in aquatic primary producer biomass. These temporal patterns among estuaries are better described as continuous gradients rather than distinct or categorical differences among classes and habitat types. The relative abundance, dominant primary producers and factors associated with their temporal patterns are discussed in detail below.

Dominant Aquatic Primary Producers. Dominance of benthic (microphytobenthos, submerged aquatic vegetation, macroalgae) over pelagic (phytoplankton) aquatic primary producers is characteristic of shallow Mediterranean estuaries such as those in the SCB (Castel *et al.* 1996, Stal *et al.* 1996, Menendez and Comin 2000, de Casabianca *et al.* 2002, McGlathery *et al.* 2007, Perez-Ruzafa *et al.* 2011). Among SCB estuaries, macroalgae was ubiquitous and most often the dominant aquatic primary producers; phytoplankton was only dominant (or co-dominant with microphytobenthos) in a handful of segments. Macroalgal biomass was dominated by green algae of the genus *Ulva spp*. These, opportunistic macroalgae species have a number of physiological traits that allow them to dominate primary producer biomass in shallow estuarine environments including rapid nutrient uptake and growth rates (Pedersen and Borum 1996, Pedersen and Borum 1997, Naldi and Viaroli 2002, Lartigue and Sherman 2005) and a high tolerance for a wide range of temperature (Fong *et al.* 1993) and salinities (Edwards *et al.* 1987, Young *et al.* 1987, Kamer and Fong 2000).

The exceptions where phytoplankton was dominant over macroalgae are noteworthy. Among segments located in large, seagrass (*Zostera* spp)-dominated estuaries, phytoplankton and microphytobenthos biomass co-dominated algal aquatic primary producers (5 of 23 estuaries). Mean annual biomass of macroalgae was roughly 15% of that found in other habitat types (intertidal and unvegetated subtidal). The second exception, Santa Clara River estuary, a bar-built estuary (SCR), was dominated throughout the year by phytoplankton and microphytobenthos, with little or no macroalgal biomass. It is reasonable to assume that intertidal and shallow subtidal areas are far more affected by macroalgal mats than deepwater habitat (>10m), simply because light available for macroalgae will limit their ability to effectively compete for nutrients in deeper water (Valiela *et al.* 1997).

The other exception to macroalgal dominance of SCB estuaries was found in those segments with ephemeral beds of *Ruppia spp.* Among the six bar-built estuaries in which the sand bar closed during the study (or remained closed), ephemeral beds of brackish submerged aquatic vegetation (dominated by *Ruppia spp.*), when present, dominated aquatic primary producer biomass in five (ZC, TC, DL, SMC, and UCL). Two of the systems with *Ruppia spp.* were continuously closed to tidal exchange throughout the study and these systems had observable biomass during every sampling period, though biomass was greatest in the late summer and fall. In the other three, *Ruppia spp.* biomass was not present during the time when the system was open and well flushed. Upon cessation of freshwater flow and inlet closure, biomass increased steadily to peak in the summer and late fall. *Ruppia spp.* was also found in three segments of estuaries open to tidal exchange. In all of these systems, there was no *Ruppia spp.* biomass during the winter months when freshwater flow was highest. These segments are notable in that they generally have a muted tidal regime, either through the presence of a tide gate, or they are located towards the head of the estuary in the more brackish zone. In these estuaries, *Ruppia spp.* may be the dominant aquatic primary producers in the segment, but not estuary, as the bed was generally limited in size.

Seasonality and Factors Associated with Aquatic Primary Producer Biomass

Sediment Characteristics. Sediment characteristics had a strong relationship with macroalgal biomass and microphytobenthos, signaling a strong, bottom up control on the relative abundance and spatial distribution of these aquatic primary producers groups (Valiela *et al.* 1997), while phytoplankton and *Ruppia spp.* biomass had no significant relationships with sediment characteristics.

Annual mean macroalgal biomass was strongly correlated with increasing %OC, %TN and %TP. This result is not surprising, given the fact that in most segments monitored, macroalgae has a strong interaction with the sediments, often found as mats in the intertidal zone or drifting just above the sediment surface in shallow subtidal waters (Thybo-Christensen *et al.* 1993, Duarte and Cebrian 1996, Kamer *et al.* 2001). In shallow, lagoonal estuaries, the feedback loop between sediments and macroalgal biomass may be one reason for it dominant status as a aquatic primary producers. Macroalgal tissue is generally very leaky, so mats can translate algal C, N and P directly to sediments (Tyler *et al.* 2003, Kamer *et al.* 2004). When macroalgae senesces, it often sinks to the bottom of the estuary where it decomposes and contributes to the carbon and nutrient pool in the sediments (Tyler *et al.* 2001, Tyler *et al.* 2003). Macroalgae also take up dissolved inorganic nutrients directly from sediments; studies have documented increased diffusive fluxes of nutrients across the sediment water interface in the presence of macroalgae because of its superior ability to draw down nutrient concentrations, thus increasing the concentration gradient and driving nutrient flux out of the sediments (Tyler *et al.* 2003, Sutula *et al.* 2006). Many sites had a spring "bloom" of macroalgae followed by a summer dip and another rise in late summer/early fall. This pattern has been observed in other estuaries (Scanlan *et al.* 2007). This may relate to availability of water-born nutrients associated with stormwater runoff in the spring, versus increased availability of remineralized nutrients from sediments, which tend to peak in late summer when sediment temperatures can peak.

With microphytobenthos, the strongest relationship was with the sediment particle size and sediment C:N ratio. microphytobenthos had a significant positive relationship with increasing sand content, and decreasing C: N ratio. Other studies have found strong linkages between microphytobenthos and sediment, which is understandable because microphytobenthos is intercalated within sediment (Henriksen *et al*. 1980, Rizzo 1990, Rysgaard *et al*. 1995, Thornton *et al*. 1999, Sundback *et al*. 2000) (Henriksen *et al.* 1980, MacIntyre *et al.* 1996, Sundbäck and Miles 2002). Billerbeck *et al*. (2007) found enhanced microphytobenthos productivity in intertidal sands relative mudflats due to higher light penetration to the microphytobenthos in sand versus fine grained sediments and more efficient transport of photosynthesis-limiting solutes to the microalgae with pore water flows in the permeable sands than in impermeable muds. Thus habitat types or estuaries with high sediment sand content will be favored for production of microphytobenthos.

Tidal Inlet Status, Class and Habitat Type. In general, the degree of tidal exchange (tidal inlet status), was a more powerful predictor of aquatic primary producer biomass then estuarine class or habitat type. The power spectra intensity of WSE on diurnal or semi-diurnal (tidal) frequencies, a continuous variable, was significantly correlated with decreasing biomass of macroalgae, phytoplankton and microphytobenthos. The diurnal power spectra intensity of WSE is a proxy for the degree of tidal flushing and residence time of water in these segments. As nutrient uptake is a rate, increased residence time serves to increase the time available for aquatic primary producers uptake of a given standing stock of nutrients in the water column (Valiela *et al.* 1997, Painting *et al.* 2007).

In contrast, tidal inlet status and estuarine class, taken as categorical variables, had no significant effect on aquatic primary producer biomass for any of the groups. Interesting, river mouth estuaries had significantly higher sand content and lower %OC and nutrient content than enclosed bays or lagoons. These continuous variables were significantly correlated with macroalgal and microphytobenthos biomass, signaling that habitat types or estuarine classes that are dominated by depositional rather than erosional processes are more likely to foster macroalgal blooms. However, the categorical variables themselves were not significant predictors, due to high variability among estuaries. For bar-built lagoons that close on a seasonal basis, this inherent susceptibility to organic matter enrichment is compounded by increased tendency to retain allochthonous or autochthonous organic matter during periods of mouth closure (Hillman *et al.* 1990).

Hydromodification, particularly the effects of anthropogenic tidal muting through diking portions of estuaries and controlled tidal flushing through tide gates, can also have a direct effect on residence time and sediment characteristics (Mitchell *et al.* 2008, Ritter *et al.* 2008). In comparison of paired segments (diked, undiked), we found that the diked segments had significantly higher phytoplankton biomass than undiked, but no significant difference was found for macroalgal biomass. This is supported by a theorem proposed by Valiela *et al*. (1997) that phytoplankton are favored over macroalgae in estuaries with longer residence times. However, we would assert that given the limited sample size of our data set, we are not able to sufficiently test for significant differences among diked and undiked sites. We suggest that a better investigation of the effects of hydromodification may lie in understanding how materials exchange and biotic connectivity is impacted through the presence of dikes and tide gates on a more estuary-specific basis.

Estuarine Water Column Nutrient Concentrations and Nutrient Ratios. In this study, we found that annually-averaged macroalgal and phytoplankton biomass had a significant correlation with estuarine water column nutrients concentrations. It is also important to note that these results reflect correlative relationships. However, viewing the correlations as potentially causal interactions, important points emerge from the analyses: 1) in the absence of scaling factors such as time averaging, time lagging, and spatial apportionment, the relationship of nutrients and chlorophyll is generally weak; 2) selecting the appropriate timescales over which to average the data is important to the outcome of the analysis; and 3) total nutrients are often better correlated with chlorophyll-a response than is dissolved inorganic nutrients as the stressor.

A number of studies have found linkages between algal response indicators and nutrient inputs for both macroalgae and phytoplankton (Kemp and Boynton 1984, Valiela *et al.* 1997, Conley *et al.* 2000, Smith 2006, Boynton and Kemp 2008). There has been some success in relating phytoplankton to both watershed nutrient loads and in-situ water column nutrient concentrations in estuaries, particularly when data are averaged over annual time periods. In general, variations in N loading rates are reflected in concentrations of N in receiving water bodies, particularly when residence time of that water body is long (on the order of weeks. Mean TN concentrations were significantly correlated to TN loading for 5 sub-systems of Chesapeake Bay averaged over a decadal period (Boynton *et al.* 2008). Conley *et al*. (2000) reported that on an annual basis about 70% of the variation in TN concentration could be explained by variation in TN loads in a large sample of Danish estuaries. Madden *et al*. (Madden *et al.* 2010) found a strong correlation between SEAWIFS remotely sensed chlorophyll-a and TN loading for 108 estuaries in the United States. In a survey of the fundamental nutrient forms and processes in several major estuaries was performed by Smith (Smith 2006) using data from 92 estuarine and coastal sites worldwide. The analysis demonstrated a strong correspondence between log transformed annual mean concentrations of total P and standing stock of chlorophyll-a and a still stronger relationship between log transformed annual mean total N and standing stock of chlorophyll-a. Nitrogen accounted for a significant portion of the variability of phytoplankton production or algal biomass on an annual basis. More in-depth analysis showed that the strength of the relationship depended on whether the nutrient data were reported as DIN concentration only ($NO_3^- + NO_2^- + NH_4^+$) or as TN (DIN + DON + PON) being generally stronger with TN than with DIN.

Nutrient concentrations are highly dynamic and are rapidly transformed by biogeochemical processing. The concentration of a dissolved inorganic nutrient measurable in the water column represents the instantaneous net “remainder” after processing by all other factors. Macroalgae have been known to take up so much N that ambient water quality in the estuary seems high, i.e., low water column nutrient concentrations and low phytoplankton biomass, even when N loads are high (Valiela *et al.* 1997, McGlathery *et al.* 2007). Measurement of particulate nutrients or total nutrients includes live planktonic biomass suspended in the water sample, so measurement of inorganic forms of N alone may underestimate the true influence of nitrogen inputs. Dissolved organic nitrogen (DON) can be the dominant form in the N pool in estuarine systems, especially during warm periods of the year when system metabolism is high. Macroalgae and phytoplankton are known to release large amounts of dissolved organic matter (Anderson and Zeutschel 1970, Valiela *et al.* 1997, Pregnall 19893) because they fix more carbon than they require and exude the unused dissolved organic carbon and nutrients, releasing up to 39% of their gross production during blooms periods (Velimirov 1986), with the remainder of fixed carbon released during senescence (Alber and Valiela 1994). In SCB estuaries, a relative large fraction of total N and P measured in each segment was dissolved organic N and P. These lines of reasoning help to explain the strong correlations between macroalgae and phytoplankton biomass and total and total dissolved nitrogen and phosphorus concentrations and N:P ratios during certain periods.

It is well established that nitrogen is the major nutrient limiting primary production in estuaries and the marine environment (Smith 1984, Hecky and Kilham 1988, Taylor *et al.* 1995, Vitousek *et al.* 1997), though phosphorus availability may affect carbon turnover by heterotrophs, ultimately limiting primary production (Thingstad *et al.* 1998, Sundareshwar *et al.* 2003). Ratios of N:P in surface waters of SCB estuaries indicate that the aquatic primary producers were, for most part, nitrogen limited. However, N:P ratios suggestive of P limitation were found in those estuaries with closed tidal inlets (SMC, ZC, SCR). This was an interesting finding, given that several of these systems were low salinity, with riverine inputs rich in effluent-, stormwater-, or ag-dominated nutrients. Other studies in similar systems have shown that peak season net primary production of macroalgae is typically limited by nitrogen supply, though N and P co-limitation and limitation by P alone can occur at certain times of the year in some cases (Fong *et al.* 1993, Peckol *et al.* 1994, Twilley 1995, Valiela *et al.* 1997, Teichberg *et al.* 2008, Teichberg *et al.* 2010).

Relationship of Aquatic Primary Producer Groups along a Gradient of Nutrient Availability. As nutrient availability increases, it has been well-documented in many parts of the world that blooms of green or red macroalgae become dominant in shallow subtidal and intertidal estuaries and lagoons, replacing seagrass or microphytobenthos (e.g., (Valiela *et al.* 1992, Peckol *et al.* 1994, Sfriso and Pavoni 1994, Hernandez *et al.* 1997, Valiela *et al.* 1997, Hauxwell *et al.* 1998, Raffaelli *et al.* 1999, Kamer *et al.* 2001, Sfriso *et al.* 2003, Viaroli *et al.* 2008). This process is referred to as a "phase shift." Under scenarios of higher nitrogen loading, Valiela *et al*. (1997) proposed that phytoplankton would eventually replace macroalgae in subtidal habitat, particularly under circumstances of higher residence time and very high nutrient concentrations. Studies documenting this transition in California estuaries are lacking. We did not observe strong evidence for this phase shift along a gradient of nutrient concentration in the 27 segment monitored. Instead, the "phase shift" appeared to be more seasonal in nature, with microphytobenthos peaking in early to late spring, while macroalgal biomass was generally low during winter and highest during the "growing season" (May-October). The strongest seasonality occurred in microphytobenthos biomass, peaking consistently across most estuaries in early to late spring. Relative to macroalgal biomass, phytoplankton biomass was generally more dominant in the winter during periods of high N concentrations. Macroalgal biomass is typically low during the winter because intermittent flood flows through estuaries effectively scour out accumulating biomass. In Santa Clara River estuary, phytoplankton blooms were chronic, reflective of year-round inputs of high N from treated POTW effluent discharges. Interestingly local managers of this estuary have noted that it can switch dominance from year to year between phytoplankton and macroalgae. Thus application of this "phase shift" concept in bar-built Mediterranean estuaries is complicated by strong confounding influences of tidal elevation, inlet closures, and interannual variability of freshwater loads.

A second way to look for evidence of a phase shift is the relative dominance of brackish submerged aquatic vegetation (*Ruppia spp.*) along a nutrient disturbance gradient. Macroalgae bloom species have the ability to shift habitat usage from benthic to floating stages, macroalgae are able to occupy all estuarine habitats by rafting in surface waters or depositing on subtidal or intertidal sediments. For this reason, brackish submerged aquatic vegetation communities, like seagrass beds, may be more vulnerable to deposition of macroalgae mats than others (e.g., Hauxwell *et al.* 2001). Likewise, phytoplankton should have the ability to outcompete submerged aquatic vegetation for light (Zaldivar *et al.* 2009). Our survey did find that macroalgae mats were found to be floating over submerged aquatic vegetation beds and macroalgae epiphytes were present on submerged aquatic vegetation stems in systems that had high macroalgae biomass (DL, UCL, SMC, LPL, SDR). However, we found no indication that high biomass of macroalgae nor of phytoplankton limited the density of *Ruppia spp.*

Several California lagoon systems are known to support very dense and apparently healthy *Ruppia spp.* populations under very eutrophic conditions (high nutrient loading, high organic loading to the sediments, fish kills, large diurnal dissolved oxygen swings, etc.. e.g., Malibu Lagoon (Sutula *et al.* 2004), Buena Vista Lagoon (McLaughlin *et al*. 2012). It is not clear if these *Ruppia spp.* beds are adapted to and thrive under high nutrient conditions or if these populations are an expression of eutrophication symptoms. In Chesapeake Bay, the growth form of seagrass and submerged aquatic vegetation are classified as “meadow forming” and “canopy forming”, respectively (Batiuk *et al.* 2001). Brackish submerged aquatic vegetation species, like *Ruppia spp.*, tend to be “canopy formers” with biomass concentrated in the top half of the water column and exhibit rapid growth toward the surface early in the growing season. Continued growth results in apical leaves near the surface of the water that actively photosynthesize, thus preventing from some degree light limitation of new growth. In contrast, “meadow forming” species, like seagrasses such as *Zostera spp.,* concentrate biomass in the lower portion of the water column and new leaf production occurs near the base of the plant, making meadow-forming species more sensitive to light limitations. In general, a better understanding is needed of the response of *Ruppia spp.* to alterations in nutrient loading, and given the limited sample size of *Ruppia spp.* dominated estuaries in our study, we view our results as not conclusive.

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IV. FACTORS AFFECTING VARIABILITY IN DISSOLVED OXYGEN CONCENTRATION IN SOUTHERN CALIFORNIA BIGHT ESTUARIES

Introduction

Dissolved oxygen (DO) is necessary to sustain the life of all aquatic organisms that depend on aerobic respiration. Eutrophication produces excess organic matter that fuels the development of low surface water DO concentrations (hypoxia) as that organic matter is respired (Diaz 2001). When the supply of oxygen from the surface waters is reduced or the consumption of oxygen exceeds the resupply (via decomposition of excessive amounts of organic matter), oxygen concentrations can decline below the limit for survival and reproduction of benthic (bottom-dwelling) or pelagic (water column dwelling) organisms (Stanley and Nixon 1992, Borsuk *et al.* 2001, Diaz 2001). Hypoxia has a number of adverse effects on aquatic organisms, including: lowered growth rates, altered behavior, reduced reproductive success, and diminished survival (Diaz and Rosenberg 1995, Breitburg *et al.* 1997, Vaquer-Sunyer and Duarte 2008). Changes in the survival and reproduction of benthic and pelagic organisms can result in habitat and biological diversity losses, foul odors and taste, and altered food webs (Sutula *et al.* 2007). Consequently, management of hypoxia in estuaries has become a global issue (Smith 1987, Karlson *et al.* 2002, Diaz and Rosenberg 2008). In cases where hypoxia has anthropogenic origins, the assumption is that hypoxia may be reduced by controlling nutrient availability and reducing the supply and/or production of oxygen-demanding organic materials to a waterbody. Thus quantifying the linkages between anthropogenic nutrient loading to estuaries, the production and respiration of allochthonous (external) and autochthonous (internal) sources of organic matter can help to provide a better understanding of how to best manage hypoxia.

Net primary production, respiration and the balance between the two, otherwise known as net ecosystem metabolism (NEM) is a useful indicator of trophic status of an estuary (Caffrey 2003a, 2004, Russell and Montagna 2007). If NEM is positive (production > respiration), the system is autotrophic and internal sources of organic matter dominate; if the NEM is negative (respiration > production), the system is heterotrophic and external sources of organic matter dominate. NEM has been suggested to be a useful indicator of eutrophication, as NEM will generally decline with increased nutrient loading into an estuary (Eyre and Ferguson 2005). As estuaries becomes increasingly eutrophic and aquatic primary producer expression shifts to dominance by phytoplankton and/or macroalgae, the large amount of labile organic matter switches the system from being net autotrophic to one where respiration largely dominates primary production (net heterotrophic).

Estuaries are highly variable in terms of physiographic setting, salinity regime, frequency and timing of freshwater flows, magnitude of tidal forcing, sediment load, stratification, residence time, and other factors (Sutula *et al*. 2011). The physical characteristics of estuarine systems have a key role in its to the effects of nutrient and organic matter loading that lead to eutrophication (Painting *et al.* 2007, Zaldivar *et al.* 2008). In particular, watershed characteristics and land use, estuarine morphology (surface area, volume, depth, etc.), hydrology and hydrodynamics, and regional climate will affect the balance of freshwater forcing and exchange at the ocean inlet, which in turn affects water retention and flushing (Painting *et al.* 2007, Zaldivar *et al.* 2008). Hypothetically, systems with a lower residence time and more flushing should experience less organic matter accumulation and consequently, be more autotrophic and well oxygenated. Thus we hypothesize that estuarine geoform (e.g., enclosed bay, lagoon and river mouth; (Madden *et al.* 2005)) as well as degree of tidal exchange with the ocean inlet (hereto referred to as tidal inlet status: open, diked, or intermittently closed) should therefore have a strong influence on NEM and extent of hypoxia in estuaries.

The purpose of this study was two- fold: 1) document differences among classes of southern California Bight estuaries and along a gradient of tidal inlet status with respect to bottom water hypoxia and measures of rates of gross primary production, respiration and NEM and 2) investigate factors affecting temporal variability in DO, including nutrient loading. This study took advantage of continuous dissolved oxygen and collateral data collected synoptically using standardized methods for 27 sites in 23 estuaries in the Southern California Bight during from January- October 2009 through the Bight Regional Monitoring Program's Estuarine Eutrophication Assessment (Section II). We make comparisons of our findings to other large and geographically diverse assessments of metabolic rates (e.g., (Caffrey 2004)).

Methods

Study Area

The Southern California Bight (SCB; Figure II-1, Section II) is an open embayment in the coast between Point Conception, California and Cabo Colnett (south of Ensenada), Baja California. The region has a Mediterranean climate, with an average annual rainfall of 10–100 cm (e.g., (Nezlin and Stein 2005)), falling primarily during winter months (December through March), and approximately 20 annual storm events (Ackerman and Weisberg 2003). Approximately 100 watersheds, encompassing fourteen thousand square miles and dominated by urban and agricultural land uses, drain into SCB estuaries and nearshore waters (Ackerman and Schiff 2003). Winter runoff to the SCB contributes more than 95% of the total annual runoff volume (Schiff *et al.* 2000, Ackerman and Weisberg 2003) and 67% of the total annual nitrogen loads (Sengupta *et al*., submitted).

SCB estuaries range in size from < 1 to > 5000 hectares. Three estuarine classes or geoforms are represented in this region (Figure IV-1): 1) enclosed bays are well flushed with a strong tidal prism and dominated by shallow or deepwater subtidal habitat. The inlet mouth is not restricted and is perennially open to tidal exchange, 2) lagoons and 3) river mouth estuaries have restricted tidal inlets, are dominated by shallow subtidal and intertidal habitat and have a long residence time due to the restricted width of the mouth. Lagoons historically received less freshwater input, though in recent years many receive substantial input from urban runoff; river mouth estuaries are dominated by fluvial forcing. For both lagoons and river mouths, the inlet can be open or closed, perennially (all year round), intermittently (open at least once per year) or ephemeral (opens infrequently, usually every several years or not known recently to open).

The conversion of open land into impervious surfaces has included dredging and filling over 75% of bays and estuaries and extensive alterations of coastal streams and rivers (Brownlie and Taylor 1981, Horn and Allen 1985, NRC 1990, Zedler 1996). These changes to hydrology alter both the timing and rate of runoff releases to coastal waters and can affect water quality through addition of sediment, toxic chemicals, pathogens, and nutrients. It has also dramatically changed the hydrology of estuaries, resulting in the type conversion from one estuarine class to another as well as fragmented sections of estuaries that are hydrologically-isolated behind levees with tidal exchange controlled by tide gates or weirs.

Figure IV-1. Examples of three major estuarine geoforms in California: enclosed bay (left), lagoon (center) and river mouth estuary (right).

Bight’08 Eutrophication Assessment Study Design

The SCB Eutrophication Assessment sought to answer the following questions: 1) what the extent and magnitude of eutrophication in SCB estuaries and 2) what the relationship between nutrient loads, estuarine nutrient concentration and indicators of estuarine eutrophication. The eutrophication assessment was conducted as a probability-based survey in which sites are randomly selected from a comprehensive list of 76 estuaries.

Because this was a regional assessment, data collection emphasized the sampling across many estuaries, rather than to better characterize eutrophication spatially within an estuary. Eutrophication is highly spatially variable within an estuary, an index area was chosen based on 1) proximity to the greatest source of freshwater nutrient loads, 2) zone in which residence time is the longest, and 3) feasibility and safety of access for frequent maintenance. This index area is hereto referred to as a "segment." Because the segment in many cases does not represent the entire estuary, the reporting unit is the estuarine segment. In each of these segments, riverine nutrient loads, primary producer abundance and DO were monitored during November 2008-October 2009.

The sampling design took into account the three subpopulations of interest: 1) estuarine geoform (enclosed bays, lagoons, and river mouth estuaries), 2) tidal regime (perennially, intermittently and ephemerally open to surface water tidal exchange) and 3) presence or absence of anthropogenic muting of tidal regime, defined by 100% containment of the segment by the presence of dikes and levees, with the presence of tide gates or weirs that reduce the amplitude of tidally-induced water level fluctuations in the estuary. These sites were selected as paired sites ("full" versus muted" within an estuary). While these were not sampled as separate strata in the survey, site selection was weighted to ensure adequate sampling of sub-populations of interest. The sample frame was developed by drawing up a comprehensive list of coastal drainages in southern California coastal watersheds and attributing these estuaries by estuarine class, tidal regime and presence of anthropogenic muting (Appendix A). Small creek mouths less than 10 m in width at the mouth and open embayments were excluded from the frame. Table II-1 (Section II) give the characteristics of the estuaries included in the assessment. A total of 27 segments were selected in 23 estuaries.

Field and Laboratory Methods

Dissolved Oxygen and Water Column Physiochemistry. Water column physiochemistry and water surface elevation was measured continuously using a YSI 6600 data sonde. Each sonde was outfitted with a conductivity/ temperature sensor, ROX optical dissolved oxygen probe, extended deployment pH probe, chlorophyll optical sensor, and a turbidity optical sensor. All sensors were treated with anti-fouling tape and calibrated at a minimum of once monthly. Sondes were deployed at one location in each segment, in bottom water (approximately 30 cm from the sediment surface). Measurements were collected every 15 minutes throughout the deployment period. Dissolved oxygen concentrations were calculated from percent saturation, temperature and salinity data. An hourly running average was applied to the data set to smooth high frequency noise.

Estuarine Water Column Nutrients and Freshwater Nutrient Loads. Within each segment a single grab sample was collected every other month for analysis for nutrients. Dissolved inorganic and total dissolved nutrients were filtered through a 0.45 μm mixed cellulose ester (MCE) filter rinsed with 20 ml of sample water (discarded) before collection into triple rinsed 30 ml HDPE sample bottles. Dissolved inorganic nutrients including ammonium (NH_4), nitrate+ nitrite (NO_3+ NO_2), and soluble reactive phosphorus (SRP) were assayed by flow injection analysis using a Lachat Instruments QuikChem 8000 autoanalyzer. Total and total dissolved nitrogen and phosphorus (TN, TP, TDN, and TDP) were via persulfate digestion followed by analysis of automated colorimetry (Alpkem or Technicon) for nitrate-N and orthophosphate-P (Koroleff 1985).

Freshwater nutrient loads were estimated using methodologies given in Sengupta *et al*. (submitted). Wet weather loads (during storm events) were measured by stormwater agencies through regular municipal stormwater National Permit Discharge Elimination System (NPDES) monitoring. Dry weather loads (during non-storm condition) were estimated from continuous flow monitoring in combination with measurement of TN and TP in every other month grab samples. Where no existing gauging of stream flow exists, water level was measured by continuous water level sensors in selected systems. Wetted channel width and velocity will be measured across the channel cross section in order to develop a rating curve for the channel.

Primary Producer Abundance. Primary producer abundance measures included macroalgae, phytoplankton, microphytobenthos (microphytobenthos) biomass.

Macroalgal abundance was determined by measuring a combination of percent cover and algal biomass in three 30 - 50 m transects in the intertidal zone at an elevation of approximately 0.3-0.6 MLLW. Percent cover was measured at ten randomly chosen points using the point-intercept method (Kennison *et al.* 2003). Biomass was collected at 5 of the quadrat locations. Each biomass sample was refrigerated until analysis and processed within 24 hours of collection. In the laboratory, algal samples were cleaned of macroscopic debris, mud and animals, and sorted to genus level. Excess water was shed from each sample, which was then weighed wet, and dried at 60°C to a constant weight, then weighed dry. During data analysis, all macroalgae genus weights were summed for each quadrat to give a total macroalgae wet and dry weight in each quadrat.

Phytoplankton biomass was estimated from fluorescence measurements collected via *in situ* optical probe (YSI 6600 sonde, chlorophyll fluorescence probe), with bi-monthly chlorophyll *a* water column grab samples taken to calibrate the continuous fluorometry. Water column chlorophyll *a* in grab samples were taken by filtering a known volume through 0.7 micron glass fiber filter and frozen until analysis using EPA method 445. *In situ* chlorophyll fluorescence was measured every 15 minutes using an optical probe mounted to a YSI 6600 V2 data sonde. Probes were maintained according to factory specifications and were routinely calibrated. Fluorescence measurements were calibrated to chlorophyll *a* concentrations using least-squares regression generated from daily averaged data probe measurements and concentration data measured in grab samples on that same day.

Sediment chlorophyll *a*, a measure of microphytobenthos, was determined on sediment composite samples collected at the beginning and end of each macroalgae transect. Composites were comprised of ten sediment plugs (3 cm in diameter, 1 cm deep) collected at each end of the transect (20 total) collected downslope of the end of each transect in approximately 30 cm of water depth. Plugs were homogenized in sample bags and refrigerated until analysis. A subsample from each bag was collected in a 15 ml centrifuge tube and frozen for benthic chlorophyll *a* analysis. Benthic chlorophyll *a* samples were analyzed for chlorophyll *a* and phaeopigments as described above for phytoplankton. The remainder was refrigerated pending analysis of grain size and sediment nutrients.

Sediment Nutrients and Grain Size. Sediment nutrients and percent fines were determined on sediment composite samples collected at the beginning and end of each macroalgae transect as described above for sediment chlorophyll *a*. Fresh sediment samples from transect composites were weighed wet and dried at 60°C to a constant weight and weighed dry. A subsample of dried sediment was ground with a mortar and pestle for analysis of percent total phosphorus (%TP), percent total nitrogen (%TN) and percent organic carbon (%OC). Samples for %OC and %TN were acidified to remove carbonates then measured by high temperature combustion on a Control Equipment Corp CEC 440HA elemental analyzer at the Marine Science Institute, Santa Barbara. Sediment %TP samples were digested via persulfate digestion then analyzed by automated colorimetry (Technicon) at the University of Georgia Analytical Chemistry Laboratory. The remainder of the sediment was reweighed dry, wet sieved through a 65μm sieve, dried at 60°C to a constant weight, and weighed dry to determine percent fines.

Data Analysis

Analysis of Tidal Inlet Forcing Using Discrete Fourier Transform (DFT). The contribution of semi-diurnal and diurnal tidal forcing regulating dissolved oxygen (DO) variability was investigated using DFT. For this, each DO time series was interpolated on regular 15-min time intervals and DFT power spectra were calculated for all estuaries for the entire period of observations using standard MATLAB routines. All spectra demonstrated two dominating peaks of variability in water surface elevation (WSE) and dissolved oxygen: diurnal (period ~1 day) and semi-diurnal (period ~0.5 day) (Figure IV-2). Diurnal periodicity can be attributed to either tidal or biological forcing. The magnitude of each peak was extracted from the DFT power spectrum (diurnal peak at 0.9–1.1 days/cycle and tidal peak at 0.4–0.6 days/cycle). Distribution of diurnal and semi-diurnal power spectra intensity among SCB estuaries was used as both a continuous variable as well as a means to categorize the estuaries with respect to tidal inlet status.

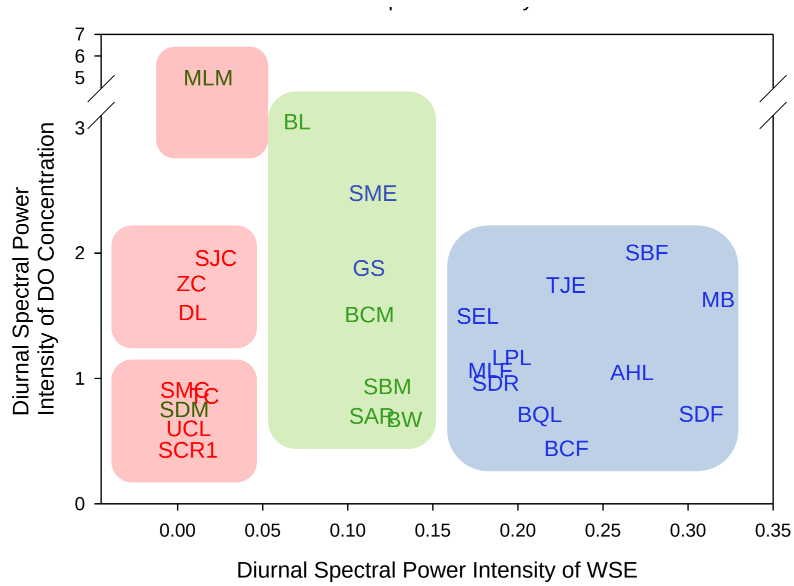

Figure IV-2. Plot of diurnal (lower panel) spectral power intensity of water surface elevation (WSE) versus dissolved oxygen concentrations. Color coding of letters designates inlet status (blue = perennially tidal, green = perennially tidal but diked, red = intermittently and ephemerally tidal). Color grouping of segments designates those that are open (blue), open but restricted (green), and severely restricted or closed (red).

Gross Primary Production, Respiration, and Net Ecosystem Metabolism. To estimate the balance between primary production and respiration (i.e., net ecosystem metabolism, or NEM) in different California estuaries, a conventional method of calculating metabolic rates from diel oxygen curve data was used (Odum 1956, Odum and Hoskins 1958). NEM is an indicator of trophic conditions within estuaries; it demonstrates whether autotrophic or heterotrophic sources of organic matter dominate. If NEM is positive, the system is autotrophic suggesting that internal production of organic matter dominates, while if NEM is negative, the system is heterotrophic and reliant on external sources of organic matter (D'Avanzo *et al.* 1996, Caffrey 2003b, Caffrey 2004). This approach is based on the assumption that oxygen is produced during daytime due to photosynthesis of autotrophic plants (phytoplankton and macroalgae), while ecosystem respiration (i.e., uptake of oxygen by aquatic animals and plants plus biochemical oxidation of autochthonous and allochthonous organic matter) occurs continuously. NEM is calculated by subtracting aerobic respiration rates from photosynthesis rates for all components contained in a defined body of water, taking into account the diffusive oxygen flux (*OF*), calculated from oxygen saturation (*DO%*). The water body is assumed to be homogenous, i.e., having the same metabolic history; in the areas where physical processes such as advection and diffusion dominate over biological processes, metabolic rates may be either underestimated or overestimated (Kemp and Boynton 1980).

First, the diffusion, or air-sea exchange (*ASE*), was estimated as follows:

$$ASE = \left(1 - \frac{DO\%(t_1) + DO\%(t_2)}{200}\right) \times 0.5 \times dt \qquad \text{(Eq. IV.1)}$$

where $DO\%(t_1)$ and $DO\%(t_2)$ are oxygen saturations (in %) for times t_1 and t_2 and *dt* is a time interval (1 hour in this study). We used a constant air-sea exchange coefficient of 0.5 g O_2 m^{-2} hr^{-1} (Caffrey 2003b), which is a good assumption at wind speeds 0–5 m s^{-1} (Russell *et al.* 2006). This coefficient was assumed to be independent of current (see Hartman and Hammond 1984) or wind velocities (see Marino and Howarth 1993). Then, *OF* (g O_2 m^{-2}) was estimated for each hourly time period as

$$OF = [DO(t_1) - DO(t_2)] \times Z - ASE \qquad \text{(Eq. IV.2)}$$

where *Z* is water depth. Oxygen fluxes during the daylight hours (from 6:00 to 18:00) were summed to give net production (g O_2 m^{-2} d^{-1}). Summed oxygen fluxes from night (18:00 to 6:00) multiplied by –1 equaled to night respiration rate. Assuming a constant respiration during the day and night, night respiration divided by hours of night equaled the hourly respiration rate (g O_2 m^{-2} h^{-1}). Total daily respiration rate (g O_2 m^{-2} d^{-1}) equaled the hourly respiration rate multiplied by 24 h. Gross production was calculated by adding net production to the hourly respiration multiplied by the daylight hours. NEM was calculated by subtracting total respiration from gross production.

This simple NEM model was based on oxygen production/consumption and diffusion and did not take into account horizontal and vertical oxygen heterogeneity and advection resulting from tidal mixing. When horizontal advection was strong, the DO variations reflected horizontal and/or vertical transport of oxygen-rich or oxygen-poor water rather than the changes in local oxygen production/consumption balance. As such the resulting time-series of oxygen production, respiration and NEM should be smoothed and analyzed over long time periods, e.g., over months or seasons.

Prevalence of Low Oxygen Concentrations. Prevalence of low oxygen concentrations in each segment on daily and seasonal time scales was estimated as the total percentage of time DO concentrations were hypoxic (< 2.8 mg O_2 L^{-1}) and oxic but low quality (% time < 5.7 mg O_2 L^{-1}) waters. These thresholds were selected to be consistent with derived acute and chronic thresholds for California estuarine organisms (Sutula 2011).

Results

Tidal Influence on DO Concentrations

The 27 estuarine segments represent a gradient of tidal influence on estuarine hydrodynamics, from strong tidal influence (diurnal spectra power intensity (SPI) >0.30), moderate (diurnal SPI of 0.10-0.30), to minor (diurnal SPI < 0.06). Not surprisingly, there was a strong influence of inlet condition on the semi- and diurnal variability in dissolved oxygen concentration (Figure IV-2). Among perennially tidal estuaries, some segments in enclosed bays (BCF, BQL, and SDF) had strong semi-diurnal components, but showed small semi-diurnal and diurnal variations in DO; in contrast, several perennially tidal bay and lagoon segments had strong diurnal variability in WSE, with higher diurnal DO variability (SDR, MLF, AHL, LPL, SEL, MB, TJE, and SBF). Notably, TJE and MLF, had strong semi-diurnal components of DO variability, but were not as distinguishable from other estuaries with respect to diurnal components.

Among the group of segments with intermediate diurnal variations in WSE (restricted), two groups were evident: 1) those surrounded by dikes and levees (SAR, BW, SBM, and BCM) with low semi-diurnal and diurnal variation in DO concentrations and 2) perennially tidal lagoons with severely restricted mouths (SME and GS).

Among those with little diurnal and semi-diurnal WSE variation (closed or extremely restricted), segments that showed little diurnal variation in DO concentration included intermittently and ephemerally estuaries (SMC, TC, UCL, SCR) and the San Diego Bay Salt pond segment (SDM). Other intermittently tidal estuaries (SJC, AC, and DL) had stronger diurnal DO variability, while two diked sites (MLM and BL) had extremely high diurnal variability with very little diurnal variation in WSE.

Variability in Carbon Metabolism and Hypoxia

Variability in dissolved oxygen concentrations was expressed using two types of measures: 1)rates of carbon metabolism, including the rates of the daily carbon production and respiration, and the net sum of these two terms- net ecosystem metabolism and 2) the prevalence of hypoxia (% time < 2.8 mg O_2 L^{-1}) and oxic but low quality (% time < 5.7 mg O_2 L^{-1}) waters.

Trends in Mean Daily Rates from Winter through Fall. Overall, the segments exhibited a great deal of variability in NEM, production, respiration, and percent of time <2.8 and 5.7 mg O_2 L^{-1} (Table IV-1, Figure IV-3). Overall, 70% of segments spent 10% of time < 5.7 mg L^{-1} and 37% of Segments spent 10 % of time < 2.8 mg L^{-1}. Sixty percent of the segments were net heterotrophic (i.e., consuming more carbon than producing through primary productivity), with NEM ranging from -1.2 ± 0.1 to -7.9 ± 0.4 g O_2 m^{-2} day^{-1}. SDR, MB, DL, TJE were noteworthy as strongly heterotrophic , with mean NEM rates from -4.4 to 7.9 g O_2 m^{-2} day^{-1}. Segments with NEM greater than -2.2 g O_2 m^{-2} day^{-1} were associated with > 9-48% of the time < 2.8 mg O_2 L^{-1} and > 25-67% of the time < 5.7 mg O_2 L^{-1}. It should be noted that MB and SDR were segments in which sondes were deployed late (May 2009), so these mean values reflect summer DO concentrations.

Table IV-1. Summary of mean and standard error of mean daily production, respiration and net ecosystem metabolism (NEM) in g O_2 m^{-2} d^{-1} and percent of time below 2.8 and 5.7 mg O_2 L^{-1} at SCB estuarine segments.

Name*		Production				Respiration				NEM			Percent of Time DO (mg L^{-1})	
		Mean	**SE**	**Mean**		**SE**	**Mean**	**SE**		**< 2.8**			**< 5.7**	
SDR		5.4	0.2	7.7		0.3	-7.9	0.3		48.0			61.9	
DL		1.6	0.4	7.5		0.3	-5.9	0.4		42.2			67	
MB		3.2	0.3	7.7		0.2	-4.5	0.3		16.0			51.9	
TJE		1.7	0.2	6.1		0.2	-4.4	0.2		15.8			45.4	
GS		0.8	0.3	4.6		0.2	-3.8	0.3		38.1			61.3	
UCL		1.2	0.3	4.9		0.2	-3.7	0.4		26.8			44.9	
SCR1		4.5	0.3	2.5		0.3	-3.1	0.5		24.9			46.6	
SEL		2.0	0.1	4.7		0.2	-2.7	0.1		7.4			26.5	
SCR2		3.5	0.6	5.8		0.5	-2.4	0.7		30.6			37.8	
ZC		2.8	0.2	5.0		0.2	-2.3	0.2		9.1			26.8	
SBF		5.3	0.2	7.4		0.2	-2.1	0.2		1.6			25.4	
BCM		3.0	0.1	5.1		0.1	-2.1	0.1		2.5			31.6	
LPL		2.5	0.2	4.4		0.2	-1.9	0.1		1.7			16.6	
SDF		1.4	0.1	3.2		0.1	-1.8	0.1		0.1			10.1	
AHL		3.1	0.1	4.8		0.2	-1.8	0.2		3			16.8	
SAR		0.8	0.1	2.1		0.1	-1.3	0.1		0			18.2	
BCF		1.4	0.1	2.6		0.1	-1.2	0.1		0			5.0	
SMR		3.7	0.2	3.9		0.2	-0.1	0.2		1.5			22.1	
SDM		1.9	0.1	1.9		0.1	0.0	0.1		0.1			8.6	
BW		2.6	0.1	2.6		0.1	0.0	0.1		59.2			64.0	
BQL		2.6	0.1	2.6		0.1	0.0	0.1		0			2.2	
MLM		4.2	0.2	4.1		0.1	0.1	0.3		16.7			38.7	
SMC		1.4	0.2	0.9		0.1	0.5	0.2		0			2.6	
SBM		1.4	0.1	0.7		0.1	0.7	0.1		0			5.8	
BL		3.6	0.1	2.6		0.1	1.1	0.2		2.4			20.8	
TPC		3.7	0.3	2.2		0.2	1.4	0.4		8.5			31.1	
MLF		2.6	0.2	0.2		0.2	2.4	0.2		0.02			1.5	

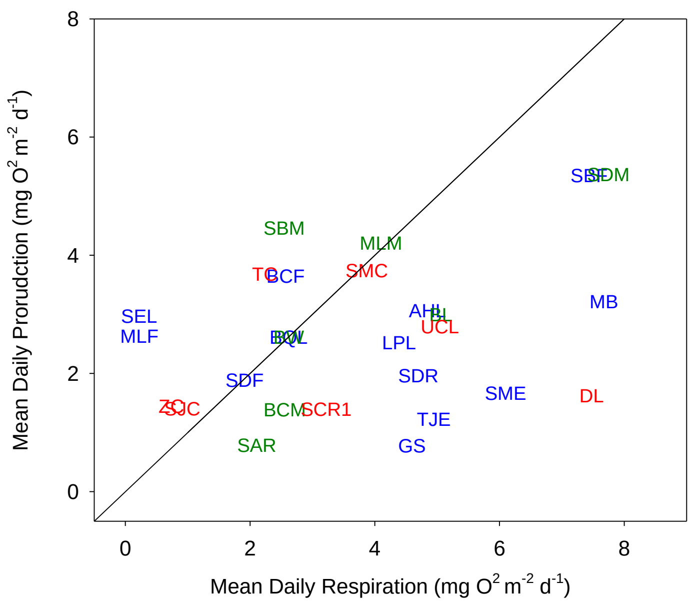

Figure IV-3. Plots of mean daily production versus respiration in the 27 estuarine segments. Line intersecting plot represents where production (P) = respiration (R). To the left of this line represents net autotrophy (P>R) and to the right represents net heterotrophy (R>P). Color coding of letters designates inlet status (blue = perennially tidal, green = perennially tidal but diked, red = intermittently and ephemerally tidal).

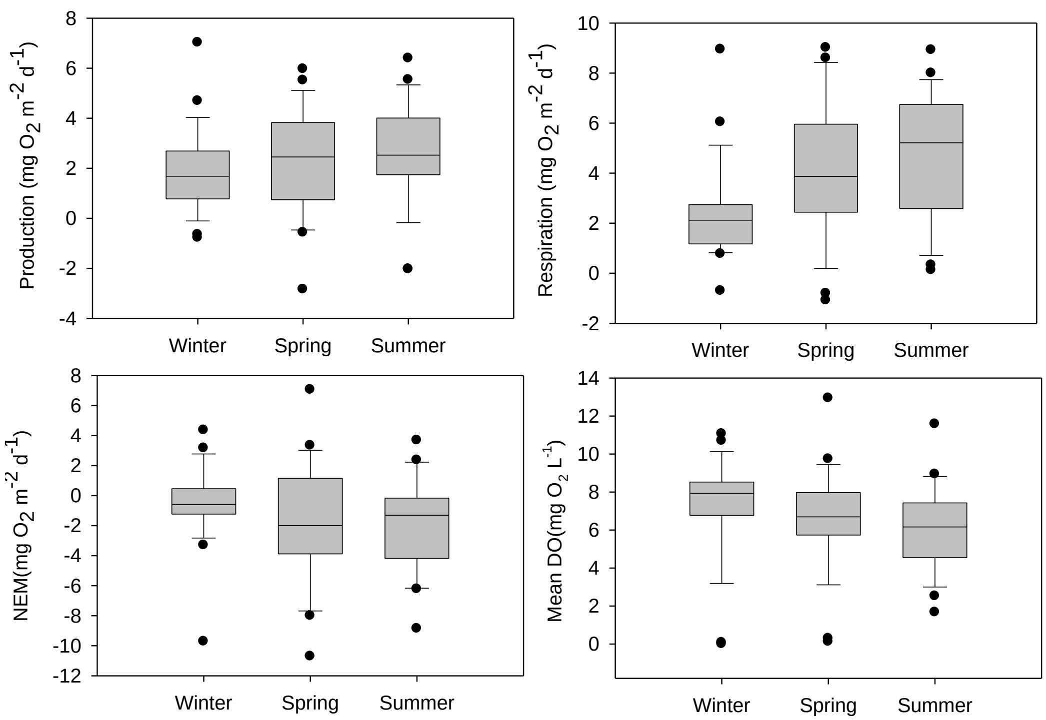

Figure IV-4. Box plots of seasonally averaged production, respiration, NEM, and dissolved oxygen across all Bight estuarine segments.

Duration of Low DO Events. High mean production and respiration rates signify a high frequency of diel (day/night) variations in DO concentration. This high frequency of strong diel variations in DO is also evident in plots of frequency and maximum duration of low DO events. Among all segments, 98% of the DO events < 5.7 mg L^{-1} occurred in durations of 24 hours or less (Figure IV-5), with the remaining 2 % lasting from 2-34 days in duration. For most perennially tidal and restricted (diked) segments, the maximum duration of any DO event < 5.7 mg l^{-1} was generally less than 24 hours and the total time spent below 5.7 mg L^{-1} was less than 30%. Perennially tidal segments TJE, AHL, MB, GS and SDR also had events longer than 24 hours in duration, ranging from 1.5 - 27.8 days (Table IV-2). All segments from intermittently and ephemerally tidal estuaries had maximum duration of low DO > 24 hours (Figure IV-6, Table IV-2).

Table IV-2. Maximum duration of DO events < 5.7 mg L^{-1} for each segment where event > 24 hours.

Estuarine Segment	Duration (days)
Perennially Tidal	
TJE	1.5
MB	6.0
AHL	8.8
GS	10.1
SDR	27.8
Perennially Tidal, Diked	
MLM	3.6
BW	6.7
Intermittently/Ephemerally Tidal	
SJC	1.5
SMC	1.5
ZC	1.9
DL	8.9
SCR	12.1
TC	19.9
UCL	36.8

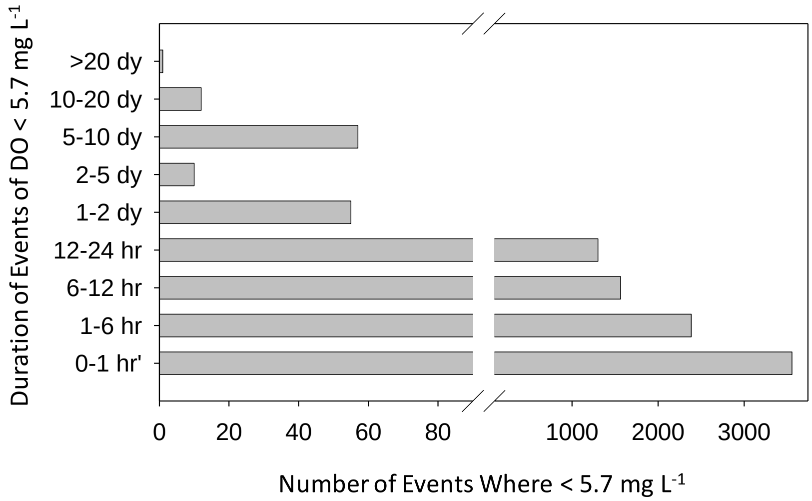

Figure IV-5. Maximum duration of DO event <5.7 mg L^{-1} found in the segment (hours) as a function of total time the segment is <5.7 mg L^{-1}.

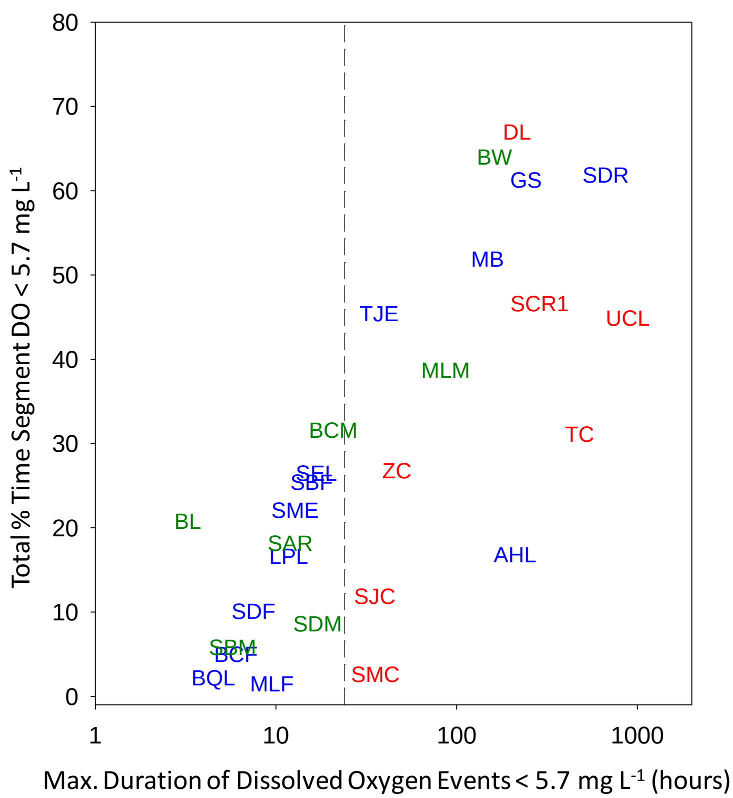

Figure IV-6. Maximum duration of DO event <5.7 mg L^{-1} found in the segment (hours) as a function of total time the segment is <5.7 mg L^{-1}. Dashed line represents 24 hours.

Effect of Estuarine Class and Inlet Status on Metabolism and Oxygen Status

No significant effect of class nor inlet status was found on NEM, respiration, production, nor percent of time DO was less than 5.7 or 2.8 mg O_2 L^{-1} (Table IV-3). In addition, we found no significant correlation between the diurnal spectral power intensity of WSE (a measure of tidal inlet status) with the production, respiration and NEM nor with extent of time in hypoxia (p-value >0.05).

Table IV-3. Results of ANOVA testing effect of inlet and class on indicators of metabolism and oxygen status.

Variable	P-Value	
	Inlet	Class
NEM	0.69	0.48
Productivity	0.44	0.07
Respiration	0.70	0.45
%Time < 2.8 mg O2 L-1	0.22	0.24
% Time < 5.7 mg O2 L-1	0.56	0.36

Environmental Factors. We investigated the correlation between a suite of environmental factors and measures of metabolism (NEM, production, respiration), status of hypoxia (total percent of time DO was less than 5.7 mg L^{-1}) and median deviation from daily running mean DO. Relationship these factors were investigated on two temporal scales:

1. Annually-averaged TN and TP loads, estuarine nutrient concentrations (TN, TP, DIN, PO4), primary producer biomass (phytoplankton, macroalgae and microphytobenthos biomass), sediment characteristics (% fines, %OC, %TN, %TP), and estuarine habitat characteristics (% of total habitat as seagrass, subtidal habitat, and intertidal habitat) across 27 estuarine segments
2. Monthly-averaged salinity, turbidity, water column chlorophyll *a*, and temperature within each estuarine segment

Across estuaries, sediment %OC, %TN and sediment C:N ratio were the only environmental factors that were significantly correlated to annually averaged NEM, respiration and % Time < 5.7 mg L^{-1} (Figure IV-7). No factors were significantly correlated with production. Annual percent of time DO less than 5.7 mg O_2 L^{-1} was positively correlated with sediment %OC and %TN. NEM and respiration was positively correlated with sediment C:N, a relationship that is counterintuitive but can be explained by the fact that sediment C:N declined with increasing sand content (Figure IV-7). Sediment %OC also had a significant positive correlation with macroalgal biomass (p-value = 0.0015, R^2 = 0.32). Dominant habitat type did not have a statistically significant effect on NEM (p-value = 0.66), though segments with brackish submerged aquatic vegetation was significantly more heterotrophic than seagrass sites (Figure IV-8).

Table IV-4 shows the results of regression analysis of monthly-averaged salinity, turbidity, water column chlorophyll *a*, and temperature with measures of metabolism and % time below 5.7 mg O^2 L^{-1}. Overall, temperature was the important factor, and generally significant in approximately half of the estuaries, depending on the variable of interest. Salinity, turbidity and chlorophyll *a* were much less important, significant only in approximately 18%, 10%, and 7% of estuaries respectively.

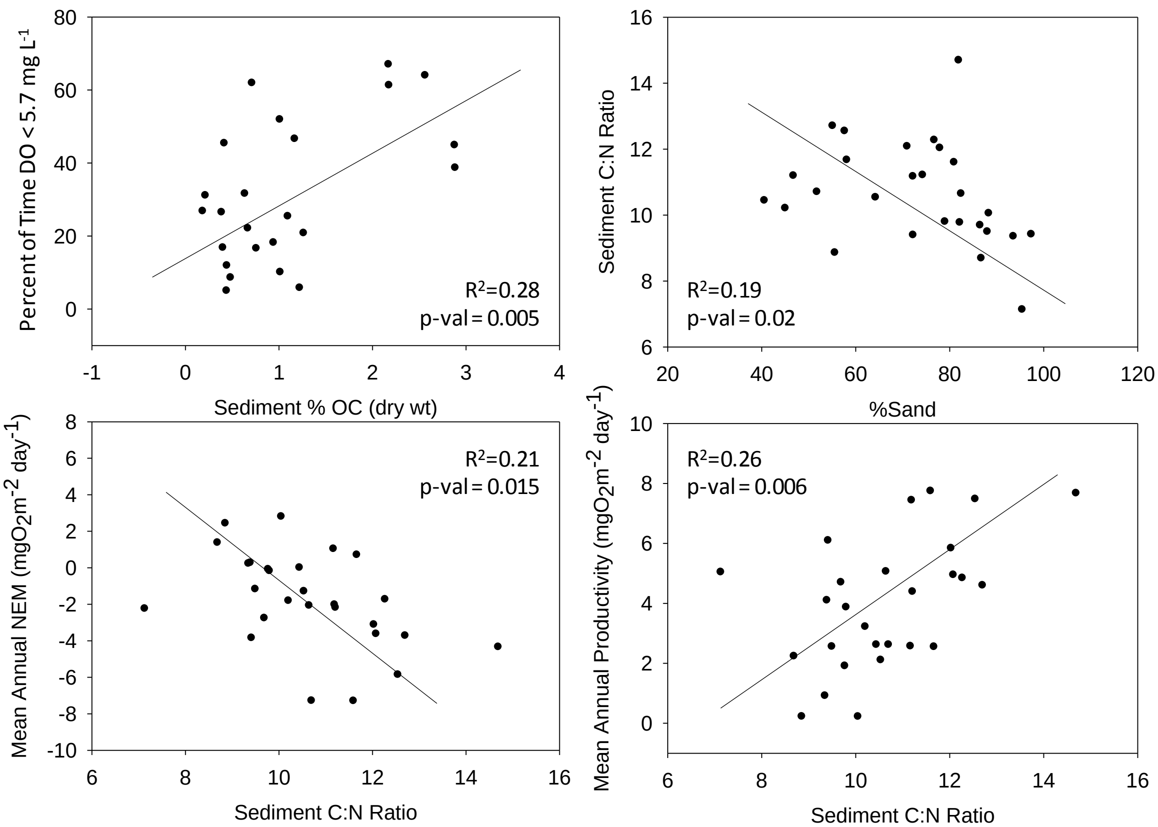

Figure IV-7. Least-squares regression relationships between sediment %OC (dry weight) and percent of time DO < 5.7 mg O_2 L^{-1} (top left panel), sediment C:N ratio and mean annual NEM and production (g O_2 m^{-2} day^{-1}; bottom left and right panel respectively), and sediment % sand and C:N ratio (top right panel).

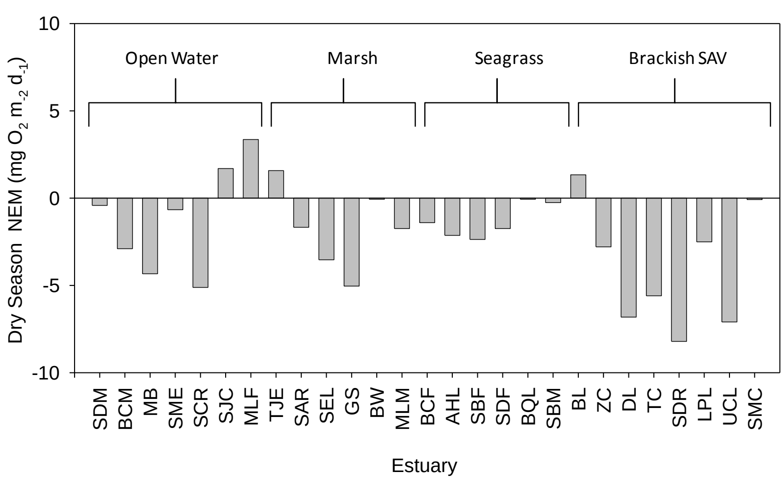

Figure IV-8. Grouping of dry season average NEM (g O^2 m^{-2} d^{-1}) by habitat type.

Table IV-4. Results of stepwise regression analysis of monthly averaged daily-averaged salinity (S), turbidity (Tu), water column chlorophyll a (C), and temperature (T) with measures of metabolism (daily NEM, production, respiration) and % time below 5.7 mg O_2 L^{-1}. NS = Not significant; "-" = not applicable.

Name	Production		Respiration		NEM		%Time Below 5.7 mg L^{-1}	
	Significant Factors	R^2	Significant Factors	R^2	Significant Factors	R^2	Significant Factors	R^2
AHL	T, S, Tu	0.81	Tu	0.38	Tu	0.87	T	0.74
BAL	T	0.41	NS	-	NS	-	Tu	0.55
BCF	T	0.54	T	0.69	T	0.58	NS	-
BCM	T	0.71	T	0.59	T	0.73	T	0.41
BQL	T	0.70	T,C	0.83	T, C	0.80	T	0.30
BW	T	0.67	NS	-	NS	-	T	0.59
DL	NS	-	T,C	0.81	NS	-	NS	-
GS	NS	-	NS	-	C	0.57	T	0.57
LPL	T	0.35	T,Tu	0.85	T	0.36	T,Tu	0.70
MB	NS	-	T, Tu, S	0.83	NS	-	T	0.39
MLF	T	0.43	NS	-	S, T	0.75	NS	-
MLM	S	0.54	T	0.80	S	0.61	T	0.59
SAR	T,S	0.66	T	0.61	S	0.53	T	0.61
SBF	C, T	0.81	T,S	0.59	C	0.54	T,S	0.81
SBM	T	0.48	T	0.73	T	0.80	NS	-
SCR1	NS	-	NS	-	T	0.67	T	0.68
SCR2	T,S	0.76	S	0.29	T, C, Tu	0.99	T	0.55
SDF	Tu, C	0.63	T	0.44	NS	0.24	T	0.43
SDM	NS	-	T	0.76	Tu	0.47	NS	-
SDR	T	0.47	T	0.51	NS	-	NS	-
SEL	S	0.50	T	0.85	T	0.46	T	0.81
SJC	C	0.42	NS	-	NS	-	NS	-
SMC	NS	-	NS	-	NS	-	T	0.37
SMR	S	0.42	T,S	0.63	T	0.31	T,S	0.42
TJE	NS	-	T	0.55	T,S	0.79	NS	-
TPC	T	0.98	NS	-	T	0.96	NS	-
UCL	Tu	0.76	S, Tu	0.84	S, Tu	0.68	T,Tu	0.91
ZC	T, Tu	0.70	T,C	0.77	Tu	0.54	T,Tu	0.70

Discussion

Comparison to Other Estuaries

Overall, NEM in SCB estuaries (-7.9 to 2.4 g O_2 m^{-2} d^{-1}) were comparable to rates estimated for other US National Estuarine Research Reserve (NERR) estuaries (-7.6 to 0.9 g O_2 m^{-2} d^{-1}; (Caffrey 2004)) and consistent with the general observation that most estuaries are heterotrophic (Smith and Hollibaugh 1993, Heip and Herman 1995, Gattuso *et al.* 1998). However, rates of production and respiration estimated by Caffrey (2004) for Pacific Coast estuaries were generally two-fold that estimated in this study, while rates for Tijuana River estuary are an order of magnitude higher that found in this study. Caffrey (2004) notes that NERRS sites were generally located off major channels in shallow regions proximal to tidal creeks and marsh, which can contribute sources of organic matter fueling higher rates of production and respiration. While this explanation seems feasible for why Caffrey (2004) respiration and production rates are larger than other previously published studies, it does not explain the discrepancy between Caffrey (2004) and this study, which are located at similar water depth and habitat types.

Sediment Organic Matter as a Driver of NEM and Extent of Hypoxia

Across SCB estuaries, a principal factor explaining variability in NEM, gross productivity, respiration and extent of hypoxia across estuaries was the mean sediment organic matter content and sediment C:N ratio. SCB estuaries are dominated by bar-built lagoons and river mouth estuaries, which generally are shallow (less than 2 m depth) and dominated by benthic primary producers. Previous studies have shown that DO dynamics in these types of estuaries are strongly regulated by the benthos, with sediment oxygen demand increasing sediments tend to accumulate organic matter (Bartoli *et al.* 1996, Viaroli and al. 1996, Eyre and Ferguson 2002, Zaldivar *et al.* 2008). Consequently, bottom waters often show large diurnal changes in oxygen concentrations associated with these high respiration rates and large sediment-oxygen demand (Sand-Jensen and Borum 1991, Viaroli and al. 1996, Viaroli *et al.* 2008). These rapid changes in benthic metabolism are enhanced by the composition of the primary producers. Our study reinforces sediment organic matter content as one of the fundamental drivers of NEM and hypoxia in estuaries; higher sediment %OC was reflected in more heterotrophic conditions and generally associated with a larger percent of time in hypoxic condition.

The significant finding of temperature as a driver for many estuary-specific models of NEM, respiration and extent of hypoxia is not surprising, given the effect of higher temperatures on biological activity in both the water column and the sediments (Caffrey 2004, Zaldivar *et al.* 2008). Sediment diagenetic processes, including organic matter decomposition, and microbially-mediated oxidation-reduction reactions such as sulfate reduction, are strongly driven by temperature (Berner 1980), and result in a larger oxygen demand as temperature increases.

Previous studies comparing NEM among estuaries have found strong relationships between NEM and estuarine nutrient concentrations, nitrogen loading and primary producer biomass (Oviatt *et al.* 1986, Hopkinson 1988, Twilley 1988, Caffrey 2004). While nutrient loading is a primary driver for eutrophication and was significantly correlated with the biomass of both macroalgae and phytoplankton in SCB estuaries (Section III), dry season NEM and extent of hypoxia were not significantly correlated to nutrient loading nor with primary producer biomass in SCB estuaries. Sediment organic matter content generally increases along a gradient of eutrophication (Nixon 1995, Pelletier *et al.* 2010), defined as the accelerated accumulation of organic matter within an estuary (Nixon 1995), but sediment organic matter accumulation is a reflection of the net accumulation of allochthonous organic matter loading and autochthonous production within an estuary over the scale of decades, rather than responding to nutrient loading on an annual time scale. This has important implications for how dissolved oxygen can be used as an indicator of eutrophication; while nutrient loads may be reduced, the recovery time required to address hypoxia problems in an estuary may be greatly extended in order for accumulated sediment organic matter to be lost from the system.

Effect of Habitat Type, Inlet Status, and Estuary Class

We hypothesized that tidal inlet status (open, diked and closed to perennial surface water exchange with the ocean) would exert a control over NEM and extent of hypoxia, as segments that were open to tidal exchange would reflect the more oxygenated ocean waters. While semi-diurnal and diurnal variability of WSE had a strong influence on diurnal variability in DO concentrations, inlet status per se was not a significant predictor on NEM nor extent of hypoxia. Comparison of paired segments within and outside of levees with tide gates or weirs showed no significant difference in rates of NEM, gross productivity, and respiration, indicating that altered hydrology alone is not an overriding factor governing NEM and oxygen status; site-specific factors such as freshwater and sediment inputs, sediment deposition and erosion, presence of legacy organic matter loading and other factors appear to be stronger drivers.

The effect of inlet status was only visible on the duration of hypoxia; the maximum duration of hypoxic event for all segments in intermittently or ephemerally tidal estuaries during "closed" inlet condition was greater than 24 hours, ranging up to 36 days. Intermittently tidal estuaries typically have density-driven stratification which develops during intermittent closure to tidal exchange when the estuaries "trap salt" and preclude diffusion and mixing of oxygen to bottom waters (Largier *et al.* 1991, Largier *et al.* 1996). Because of this salt trap effect, intermittently tidal estuaries are prone to "natural" bottom-water hypoxia (Largier *et al.* 1997, Gaines *et al.* 2006), a condition that is exacerbated during eutrophication.

Habitat type was also hypothesized to be an important driver. Caffrey (2004) found that habitat type was a significant predictor of NEM in 42 sites from 22 US NERR estuaries; NEM in seagrass-dominated or macroalgal-dominated habitat types were generally balanced or autotrophic, while open water, marsh and mangrove-dominated sites were net heterotrophic. In our study, we found no significant effect habitat type on NEM and seasonal extent of hypoxia in SCB estuaries. NEM in seagrass- and brackish submerged aquatic vegetation dominated segments of SCB estuaries was net heterotrophic, while open water and marsh-dominated segments were variable, from net autotrophic to net heterotrophic. Sediment organic content in SCB estuarine segments had significant positive correlation to percent intertidal habitat, from high content in tidal channels, marsh and intertidal flats to lower content in main channels or open water areas where tidal scour is higher (Caffrey 2004). Gradients of sediment organic matter content and other factors controlling NEM and hypoxia occur across estuarine classes and habitat types, thus obscuring or overriding the importance of these categorical variables like habitat types and class.

Interestingly NEM of seagrass-dominated (*Zostera sp*.) segments appears to be distinct, if not statistically different, from segments dominated by brackish submerged aquatic vegetation, typically dominated by *Ruppia spp.* Six of eight brackish submerged aquatic vegetation-dominated estuaries were strongly heterotrophic. Populations of *Ruppia spp.* found in these segments are canopy forming (Batuik *et al.* 2000, Sutula 2011), where the biomass is concentrated in the top half of the water column and exhibit rapid growth toward the surface early in the growing season. Canopy formation results in shading of older portions, the sloughing of lower leaves and accumulation of degraded organic matter at the sediment. When dense canopies form, DO stratification is frequently observed (Sutula *et al.* 2010a). In contrast, "meadow forming" species such as *Zostera spp*. concentrate biomass in the lower portion of the water column and new leaf production occurs near the base of the plant. These basic differences between canopy- and meadow-forming submerged aquatic vegetation species may be contributing to differences in NEM among these segments. Another factor to consider is that six of eight estuaries dominated by *Ruppia spp.* are intermittently tidal estuaries. As discussed above, these estuaries are prone to strong density driven stratification as dense salty water is trapped behind the sand berm. The strongly heterotrophic conditions in these intermittently tidal estuaries may be driven by this 'salt trap' effect (Largier *et al.* 1991, Largier *et al.* 1996), but further exacerbated by the presence of dense canopy forming submerged aquatic vegetation which further restrict recirculation and re-oxygenation of the water column (Sutula *et al.* 2010b). Additional work is needed to better characterize the dissolved oxygen budgets of brackish-submerged aquatic vegetation dominated estuaries, with linkage to basic physiological requirements, environmental triggers to seasonal cycles of submerged aquatic vegetation growth, as well as the seasonal cycles connectivity to the ocean.

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V. INVESTIGATION OF THE RELATIONSHIP BETWEEN NUTRIENT INPUTS AND EXTENT OF EUTROPHICATION

Introduction

Eutrophication, defined as the accelerated increase of organic matter accumulation (Nixon 1995), is a global environmental issue (Nienhuis 1992, Jorgensen and Richardson 1996, Valiela and Bowen 2002, Kemp *et al.* 2005, Paerl *et al.* 2006, Zaldivar *et al.* 2008, Duarte 2009, Garmendia *et al.* 2012). The ecological impacts of eutrophication on coastal waters can have far-reaching consequences, including fish-kills and lowered fishery production (Glasgow and Burkholder, 2000), degradation of seagrass and kelp beds (Twilley 1985, Burkholder *et al.* 1992, McGlathery 2001), smothering of benthic organisms (Rabalais and Harper 1992), nuisance odors, and impacts on human and marine mammal health from increased harmful algal blooms and poor water quality (Bates *et al.* 1989, Bates *et al.* 1991, Trainer *et al.* 2002). According to EPA, eutrophication is one of the top three leading causes of impairments of the nation's waters (USEPA 2000); the most recent NOAA National Estuarine Eutrophication Assessment (NEAA) found that the majority of estuaries assessed had overall eutrophic conditions rated as moderate to high and, on the whole, eutrophication in US estuaries was increasing (Bricker *et al.* 2007).

Eutrophication in estuaries is strongly linked to an increase in nutrient and organic matter inputs (Pinckney *et al.* 2001), with rivers as the primary source (Maybeck 1982). In some urbanized regions, riverine nutrient inputs have increased as much as 20-fold from pre-industrial times, associated with increasing population and associated urbanization of watershed land use (Howarth *et al.* 1996). Estuaries show a differential response to nutrient inputs, in part because of the range in lag time between the load and response variables (e.g., algal biomass, dissolved oxygen) as well as other factors that mitigate response to nutrient inputs such as physiographic setting, salinity regime, frequency and timing of freshwater flows, magnitude of tidal forcing, sediment load, stratification, residence time, denitrification, and other factors (Kemp and Boynton 1984, Malone *et al.* 1988, Dettmann 2001, Pinckney *et al.* 2001). This combination of factors results in differences in the dominant primary producer communities as well as variability in the pathways that control how nutrients cycle within the estuary. Nixon *et al*. (Nixon and al. 1996) and Dettman (Dettmann 2001) have demonstrated that the regression relationship between nutrient inputs and response improves by taking in to account factors such as freshwater residence time, estuarine volume, and denitrification rate.

There has been a great deal of discussion among scientists and managers about whether estuarine surface water nutrient concentrations or nutrient loading to the estuary is a more relevant as target for management of eutrophication. If nutrient loads are the focus, management may focus on source reduction, capture and treatment of loading from storm events, which have lower concentrations than dry weather flow but higher flows and thus can represent the majority of annual nutrient loads into southern California estuaries (Ackerman and Schiff 2003), Sengupta *et al*. submitted). If concentrations are the focus, regulation of dry weather inputs, which have higher nutrient concentrations but lower flow, may be of higher priority, particularly in Mediterranean climates where dry weather conditions constitute the majority of time during an annual cycle (Cushing *et al.* 1995).

Nutrient loads, which represent the source of nutrients to the system during a given time period, has been advocated as a more integrated measure of the magnitude of ecosystem-level primary producer response from a mass balance perspective. In contrast, ambient nutrient concentrations are a measure what is available for primary producer uptake on short timescales (e.g., phytoplankton, macroalgae, vascular plants, etc.) (Boynton and Kemp 2000). For example, macroalgae are known to rapidly take up dissolved inorganic nitrogen and, over timescales of hours to days, can bring surface water concentrations down to near detection limits; a study of five estuaries in macroalgal dominated estuaries found no relationship with estuarine nutrient concentrations (Kennison *et al.* 2003). Thus, depending on the time scale of nutrient delivery to the estuary versus biological response, ambient nutrient concentrations often reflect the remaining inventory of nutrients that are left over or that have been recycled into organic forms. Scientists and environmental managers must understand how ecological response indicators such as algal biomass are linked to nutrient inputs if they are to properly design programs to limit eutrophication and set effective water quality goals for restoration and preservation (Dettmann 2001).

The purpose of this study was to investigate the relative strength of the empirical relationships between symptoms of eutrophication (i.e., primary producer abundance) and estuarine nutrient concentrations or watershed nutrient loads. For this purpose, we utilized a one -year synoptic data set of estuaries during the Southern California Bight Regional Monitoring Program (RMP) estuarine eutrophication assessment. The SCB estuarine eutrophication assessment consisted of collection of data on response indicators and watershed nutrient loading in 27 segments in 23 estuaries from Santa Barbara to the U.S. border with Mexico from November 2008 through October 2009. We used statistical models to determine the strength of the relationships between extent of eutrophication and nutrient concentrations or loads.

Methods

Study Area

The Southern California Bight (SCB; Figure II-1, Section II) is an open embayment in the coast between Point Conception, California and Cabo Colnett (south of Ensenada), Baja California. The region has a Mediterranean climate, with an average annual rainfall of 10–100 cm (e.g., (Nezlin and Stein 2005)), falling primarily during winter months (December through March), and approximately 20 annual storm events (Ackerman and Weisberg 2003). Approximately 100 watersheds, encompassing fourteen thousand square miles and dominated by urban and agricultural land uses, drain into SCB estuaries and nearshore waters (Ackerman and Schiff 2003). Winter runoff to the SCB contributes95% of the total annual runoff volume (Schiff *et al.* 2000, Ackerman and Weisberg 2003) and 67% of the total annual nitrogen and phosphorus loads.

SCB estuaries range in size from less than 1 to greater than 5000 hectares. Three estuarine classes or geoforms are represented in this region: 1) enclosed bays have unrestricted ocean inlets and are perennially open to tidal exchange, well flushed with a strong tidal prism and dominated by shallow or deepwater subtidal habitat, 2) lagoons and 3) river mouth estuaries both have restricted tidal inlets, are dominated by shallow subtidal and intertidal habitat and have a long residence time due to the restricted width of the mouth. Lagoons historically received less freshwater input, though in recent years many receive substantial input from urban runoff; river mouth estuaries are dominated by fluvial forcing. For both lagoons and river mouths, the inlet can be open or closed, perennially (all year round), intermittently (open at least once per year) or ephemeral (opens infrequently, usually every several years or not known recently to open).

The conversion of open land into impervious surfaces has included dredging and filling over 75% of bays and estuaries and extensive alterations of coastal streams and rivers (Brownlie and Taylor 1981, Horn and Allen 1985, NRC 1990, Zedler 1996). These changes to hydrology alter both the timing and rate of runoff releases to coastal waters and can affect water quality through addition of sediment, toxic chemicals, pathogens, and nutrients. It has also dramatically changed the hydrology of estuaries, resulting in the type conversion from one estuarine class to another as well as fragmented sections of estuaries that are hydrologically-isolated behind levees with tidal exchange controlled by tide gates or weirs.

Bight'08 Eutrophication Assessment Study Design

The SCB Eutrophication Assessment sought to answer the following questions: 1) what the extent and magnitude of eutrophication in SCB estuaries and 2) what the relationship between nutrient loads, estuarine nutrient concentration and indicators of estuarine eutrophication. The eutrophication assessment was conducted as a probability-based survey in which sites are randomly selected from a comprehensive list of 76 estuaries.

Because this was a regional assessment, data collection emphasized the sampling across many estuaries, rather than to better characterize eutrophication spatially within an estuary. Eutrophication is highly spatially variable within an estuary, an index area was chosen based on 1) proximity to the greatest source of freshwater nutrient loads, 2) zone in which residence time is the longest, and 3) feasibility and safety of access for frequent maintenance. This index area is hereto referred to as a "segment." Because the segment in many cases does not represent the entire estuary, the reporting unit is the estuarine segment. In each of these segments, riverine nutrient loads, primary producer abundance and DO were monitored during November 2008-October 2009.

Site selection was conducted by drawing up a comprehensive list of coastal drainages in southern California coastal watersheds and attributing these estuaries by estuarine class, tidal regime and presence of anthropogenic muting (Appendix A). Small creek mouths less than 10 m in width at the mouth and open embayments were excluded from the frame. Table II-1 (Section II) gives the characteristics of the estuaries included in the assessment. A total of 27 segments were selected in 23 estuaries.

Field and Laboratory Methods

Macroalgae Abundance. Monitoring of macroalgal abundance provided information on when algal blooms occur in each class of estuary, how far they extend spatially, and how long they endure.

Macroalgal abundance was determined by measuring percent cover and algal biomass. Within each index area, three 30 - 50 m transects were laid out in the intertidal area, parallel to the water's edge and along the same elevational contour at approximately three quarters of the distance from the mean lowest low water line to the downslope end of vascular vegetation on the mid-to-upper mudflat. This area has been demonstrated to be representative of macroalgae accumulation in southern California estuaries (Kennison *et al.* 2003). Percent cover was measured at ten randomly chosen points along each transect by placing a 0.5 m^2 quadrat with 49 intercepts on the benthos and recording the presence or absence of each macroalgae species under each intercept. Biomass was collected at 5 of the quadrat locations. Each biomass sample was refrigerated until analysis and processed within 24 hours of collection. In the laboratory, algal samples were cleaned of macroscopic debris, mud and animals, and sorted to genus level. Excess water was shed from each sample, which was then weighed wet, and dried at 60°C to a constant weight, then weighed dry. During data analysis, all macroalgae genus weights were summed for each quadrat to give a total macroalgae wet and dry weight in each quadrat. Because of lack of confidence in taxonomic expertise among the field groups, a decision was made to lump macroalgal biomass and cover data into broad taxonomic groups (green, red), maintaining wrack separately.

Phytoplankton Biomass. Phytoplankton biomass was estimated from fluorescence measurements collected via *in situ* optical probe (YSI 6600 sonde, chlorophyll fluorescence probe), with bi-monthly discrete chlorophyll *a* water samples taken to calibrate the continuous fluorometry. Discrete suspended chlorophyll *a* pigments were concentrated from 250-500 ml of sample water by filtering at low vacuum through a 45 mm diameter Whatman glass fiber filter. Filters were stored in a petri-dish covered in aluminum foil and frozen until analysis. Photosynthetic pigments were extracted from filters in 90% acetone solution and allowed to steep overnight, to ensure complete extraction of chlorophyll *a* (EPA 445). Fluorescence was measured before and after acidification with 0.1 M HCl to determine the phaeophytin-corrected chlorophyll *a*. Concentrations were calculated relative to a laboratory standard. *In situ* chlorophyll fluorescence was measured every 15 minutes using an optical probe mounted to a YSI 6600 V2 data sonde. Probes were maintained according to factory specifications and were routinely calibrated. Fluorescence measurements were calibrated to chlorophyll *a* concentrations using least-squares regression generated from daily averaged data probe measurements and discrete concentration data collected on that same day.

Dissolved Oxygen and Water Column Physio-chemistry. Water column physio-chemistry was measured continuously using a YSI 6600 data sonde. Each sonde was outfitted with a conductivity/ temperature sensor, ROX optical dissolved oxygen probe, extended deployment pH probe, chlorophyll optical sensor, and a turbidity optical sensor. All sensors were treated with anti-fouling tape and calibrated at a minimum of once monthly. Sondes were deployed at one location in each segment, in bottom water (approximately 30 cm from the sediment surface). Measurements were collected every 15 minutes throughout the deployment period. Dissolved oxygen concentrations were calculated from percent saturation, temperature and salinity data. An hourly running average was applied to the data set to smooth high frequency noise.

Estuarine Water Column Nutrients and Freshwater Nutrient Loads. Within each segment a single grab sample was collected every other month for analysis for nutrients. Dissolved inorganic and total dissolved nutrients were filtered through a 0.45 μm mixed cellulose ester (MCE) filter rinsed with 20 ml of sample water (discarded) before collection into triple rinsed 30 ml HDPE sample bottles. Dissolved inorganic nutrients including ammonium (NH_4), nitrate+ nitrite (NO_3+ NO_2), and soluble reactive phosphorus (SRP) were assayed by flow injection analysis using a Lachat Instruments QuikChem 8000 autoanalyzer. Total and total dissolved nitrogen and phosphorus (TN, TP, TDN, and TDP) were via persulfate digestion followed by analysis of automated colorimetry (Alpkem or Technicon) for nitrate-N and orthophosphate-P (Koroleff 1985).

Freshwater nutrient loads were estimated using methodologies given in Sengupta *et al*. (submitted). Wet weather loads (during storm events) were measured by stormwater agencies through regular municipal stormwater National Permit Discharge Elimination System (NPDES) monitoring. Dry weather loads (during non-storm condition) were estimated from continuous flow monitoring in combination with measurement of TN and TP in every other month grab samples. Where no existing gauging of stream flow exists, water level was measured by continuous water level sensors in selected systems. Wetted channel width and velocity will be measured across the channel cross section in order to develop a rating curve for the channel.

Data Analysis

Estuarine Characteristics. Nutrient loads were normalized by estuarine area, volume and residence time. Estuarine area and volume data were gathered from two sources: 1) summarized estuarine area associated with specific estuarine habitat types, generated from National Wetland Inventory wetland maps (www.socalwetlands.org) and seagrass maps (Bernstein *et al*. 2011), 2) merged bathymetry/ topography data layer generated from LiDAR and existing bathymetric data as a part of a study of estuarine classification (Sutula *et al*., unpublished data). Residence time was estimated using a combination of previously published estimates, the bathtub model approach for estuaries with closed inlet, and unpublished rates from previous hydrodynamic modeling.

Timescales. Statistical relationships between primary producer communities and nutrient inputs were assessed over a variety of timescales: annual (water year: Nov 2008- Oct 2009), wet season/winter (Nov 08- Apr 09), dry season/summer (May 09-Oct 09), wet weather (during a storm event), dry weather (all other times excluding storm events). Macroalgae and phytoplankton biomass represent the average of all measurements taken within the timeframe for each segment; macroalgae is an average of the discrete segment biomass and cover values during sampling periods and phytoplankton is the average of all water column chlorophyll *a* measurements measured with the in situ data sonde. Peak season macroalgae is defined as the average biomass/cover of segment values for the two consecutive periods of highest biomass/cover. The "maximum" period is defined as the highest single period of biomass/cover. Phytoplankton 90th percentile is a measure of the bloom concentration of suspended chlorophyll *a*. Means, standard errors, population maximum, and population minimums were all calculated using MS Excel.

Nutrient Forms. Water column nutrient concentrations were partitioned into nutrient species. Particulate nitrogen and phosphorus (PN and PP) was calculated from the difference between the total fraction (TN orTP) and the total dissolved fraction (TDN orTDP). Dissolved organic nitrogen and phosphorus (DON and DOP) is calculated from the difference of the total dissolved fraction (TDN orTDP) minus the dissolved inorganic fraction (DIN: $NH_4 + NO_3 + NO_2$; DIP = ortho-PO_4). Data on nutrient load partitioned by nutrient species (particulate phase, dissolved organic phase, dissolved inorganic phase) was only available for one sampling event, so analysis of relationships with other variables were limited to TN and TP loads.

Statistical Analysis. Relationships between macroalgal and phytoplankton biomass and watershed nutrient loads and estuarine water column nutrient concentations were investigated using least-square s regressions on log-transformed data. Statistical tests were conducted on JMP 9.0 (SAS Institute Inc.).

Results

Riverine Nutrient Loads and Segment Water Column Concentrations

Total nitrogen loads to SCB estuaries ranged from 190 to 350,000 kg yr^{-1} TN, while total phosphorus loads ranged from 25 to 83,000 kg yr^{-1} TP (Figure V-1, Table V-1). Normalizing loads over estuarine area (areal loads) yielded a range from 2 to 465 g m-2 yr^{-1} TN and 0.02 to 120 g m-2 yr^{-1} TP (Figure V-1). On average, 84% of the TN and 85% of the TP loads were delivered during wet weather.

Annual average nutrient concentration range from 8.9 – 3260 μM TN and 1.7 μM to 33 μM TP (Figure V-2). TN concentrations were not significantly different between winter and summer dry weather, though significant differences were evidenced among dominant nutrient form. Mean DIN was significantly higher during winter compared to summer dry weather ($p = 0.0008$), while DON was significantly higher in summer compared to winter dry weather ($p = 0.0162$). PN concentration was not significantly different between summer and winter. TP concentrations were significantly higher during summer compared to winter dry weather ($p = 0.0010$), and the composition of the phosphorus pool was also significantly different between seasons. SRP, DOP, and PP were all higher during summer compared to winter dry weather ($p = 0.0203$, $p = 0.0363$, $p = 0.0042$, respectively).

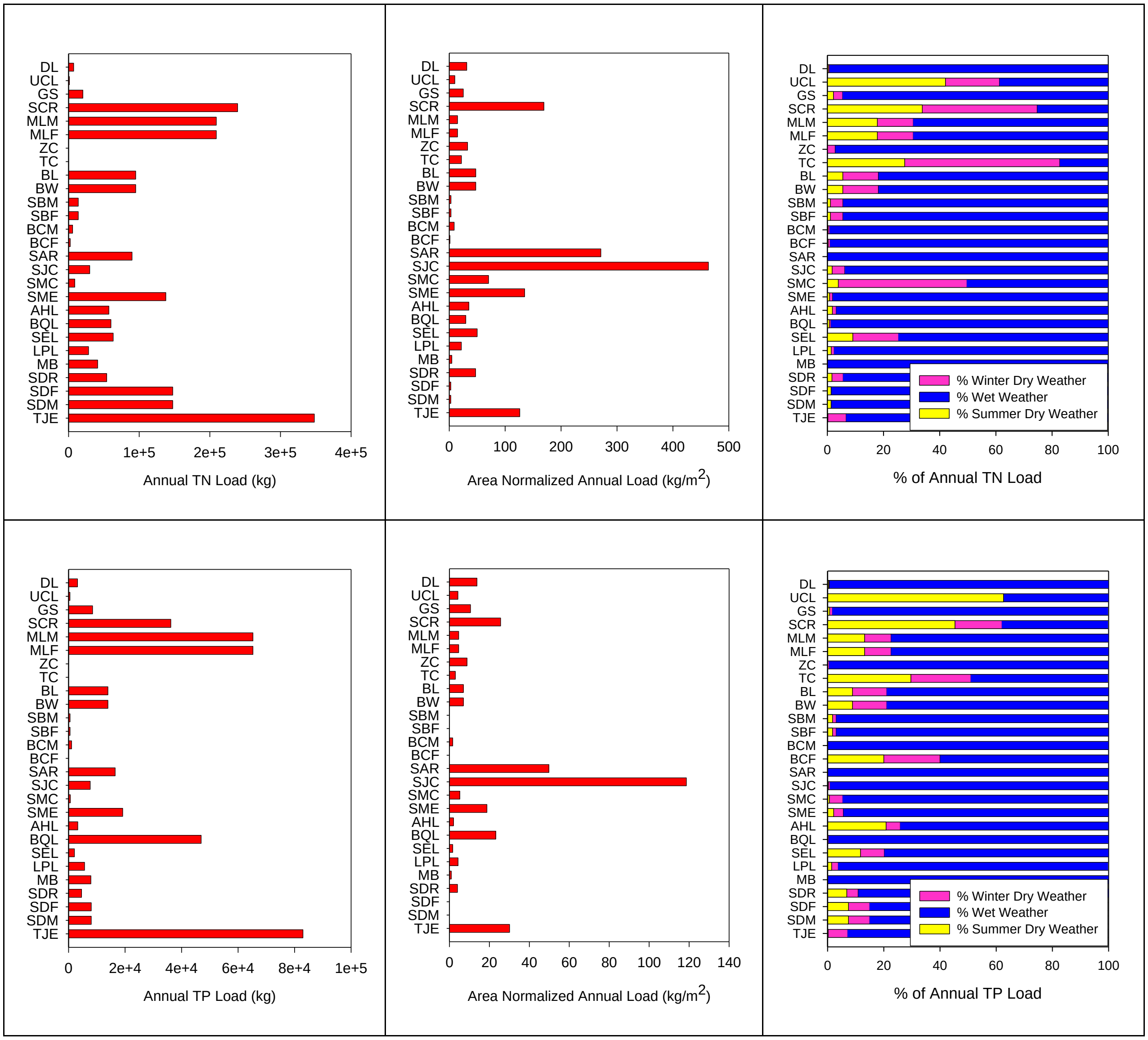

Figure V-1. TN and TP annual loads, area normalized TN and TP loads, and percent of annual load attributable to wet weather, winter dry weather and summer dry weather flow into SCB estuaries.

Table V-1. Estuarine area, volume, residence time, TN and TP loads.

Segment	Estuarine Area (km^2)	Estuarine Volume (km^3)	Dry Season Residence Time (days)	Annual TN Load (kg)	Wet Weather TN Load (kg)	Dry Weather TN Load (kg)	Dry Season TN Load (kg)	Annual TP Load (kg)	Wet Weather TP Load (kg)	Dry Weather TP Load (kg)	Dry Season TP Load (kg)
DL	231,885	11,847	140	7,287	7,230	57	27	3,198	3,177	21	13
UCL	124,229	107,595	25	1,257	487	771	529	532	310	222	333
GS	804,812	946,089	5	20,369	19,258	1,112	446	8,518	8,372	146	65
SCR	1,412,587	1,381,177	40	239,378	60,485	178,893	80,977	36,212	13,730	22,483	16,428
MLM	14,098,340	5,637,426	80	209,210	145,172	64,038	37,278	65,281	50,512	14,769	8,598
MLF			3	209,210	145,172	64,038	37,278	65,281	50,512	14,769	8,598
ZC	17,544	4,873	40	578	562	16	0	154	153	1	0
TC	8,498	1,851	3	187	32	155	51	25	12	13	8
BL	1,996,200	7,763	15	95,098	77,814	17,284	5,246	13,939	10,998	2,940	1,239
BW	1,996,200	242,036	5	95,098	77,814	17,284	5,246	13,939	10,998	2,940	1,239
SBM	4,194,998	8,033,027	40	13,832	13,064	768	155	550	534	17	10
SBF			15	13,832	13,064	768	155	550	534	17	10
BCM	651,724	983,671	7	5,840	5,790	50	17	1,081	1,079	2	0
BCF	1,708,457	3,046,792	3	2,500	2,475	25	8	25	15	10	5
SAR	331,670	201,881	5	89,981	89,975	6	0	16,528	16,527	1	0
SJC	64,839	60,820	50	30,062	28,196	1,867	513	7,695	7,623	71	22
SMC	126,262	19,465	15	8,895	4,474	4,422	347	656	621	36	4
SME	1,020,806	2,127,854	5	137,799	135,184	2,616	1,122	19,177	18,101	1,076	419
AHL	1,618,742	3,985,366	3	57,185	55,368	1,817	1,033	3,283	2,433	850	683
BQL	2,022,063	4,845,723	3	60,037	59,267	770	438	46,966	46,768	198	52
SEL	1,261,824	479,545	5	63,262	47,211	16,051	5,746	2,080	1,660	420	243
LPL	1,306,657	1,531,555	3	28,325	27,631	694	394	5,669	5,453	216	83
MB	8,795,281	47,367,098	10	41,283	41,250	33	7	7,902	7,896	6	2
SDR	1,142,328	1,940,952	3	54,066	51,012	3,053	889	4,629	4,125	504	317
SDF	50,094,111	339,772,077	40	147,537	145,548	1,989	1,989	8,045	6,838	1,207	600
SDM			65	147,537	145,548	1,989	1,989	8,045	6,838	1,207	600
TJE	2,755,819	3,269,973	3	348,018	324,662	23,355	741	82,988	77,024	5,964	189

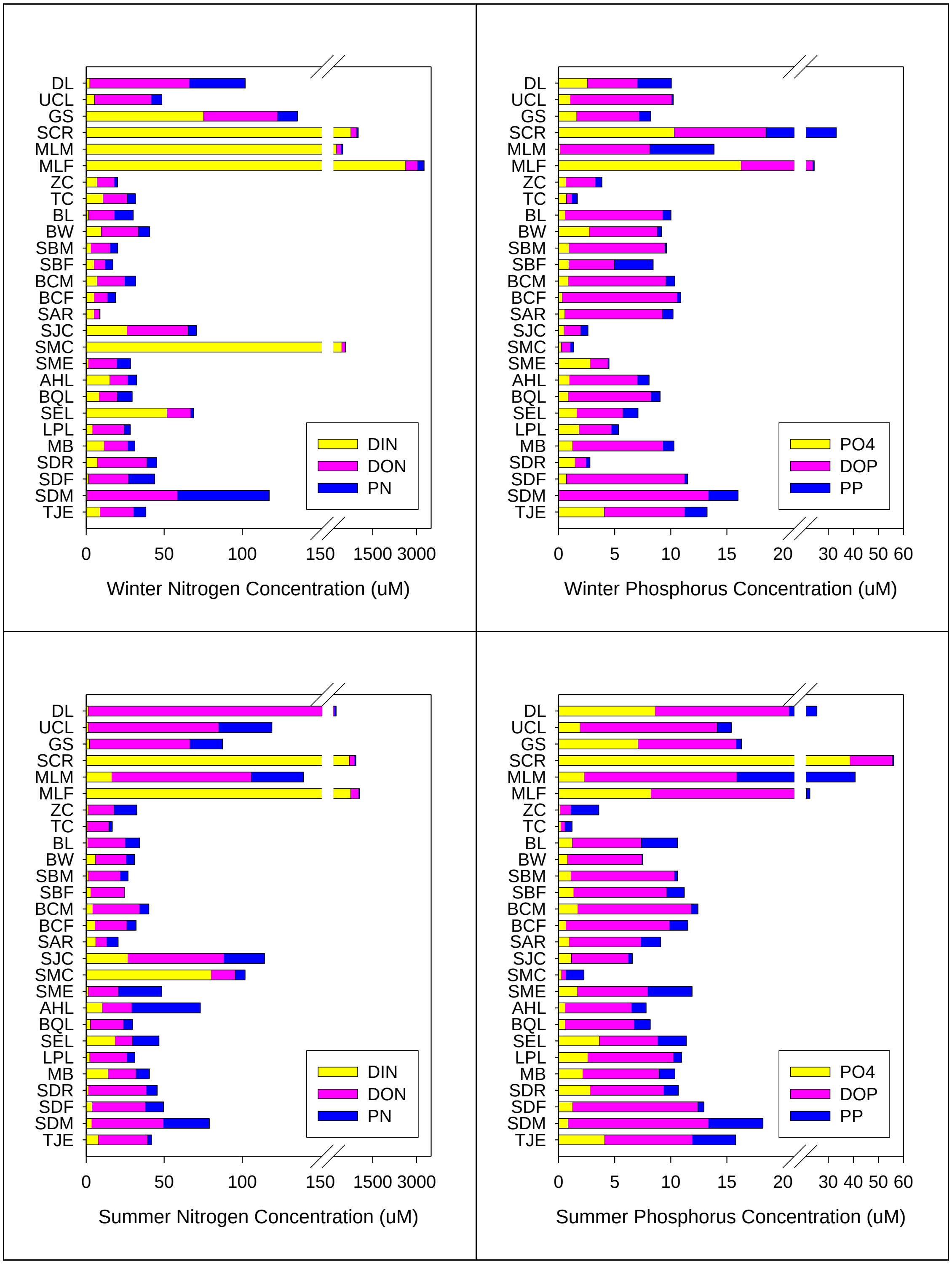

Figure V-2. Composition of mean winter and summer dry weather TN and TP concentration by form in the segment surface waters.

Relationship Between Watershed Nutrient Loads and Estuarine Water Column Nutrients

Watershed nutrient loads had a significant positive correlation to estuarine water column nutrient concentrations, though the strength of the least-square regression relationships varied depending on averaging period and constituent. Relationships were investigated on four timescales: annual, wet weather (storm events), dry weather (annual excluding storm events), and dry season (summer). On an annual timescale, annual average water column nutrients were had a significant, positive correlation with annual TP loads ($R^2 = 0.39$) but not TN loads ($R^2 = 0.07$) (Figure V-3A and 4B). Wet season TN loads and TP loads were not significantly related to wet season water column nutrient concentrations. Dry season TN and TP loads had a significant positive relationship with mean dry season TN and TP concentrations at the segment site ($R^2 = 0.29$ for TN and $R^2 = 0.30$ for TP; Figure V-3C and 3D).

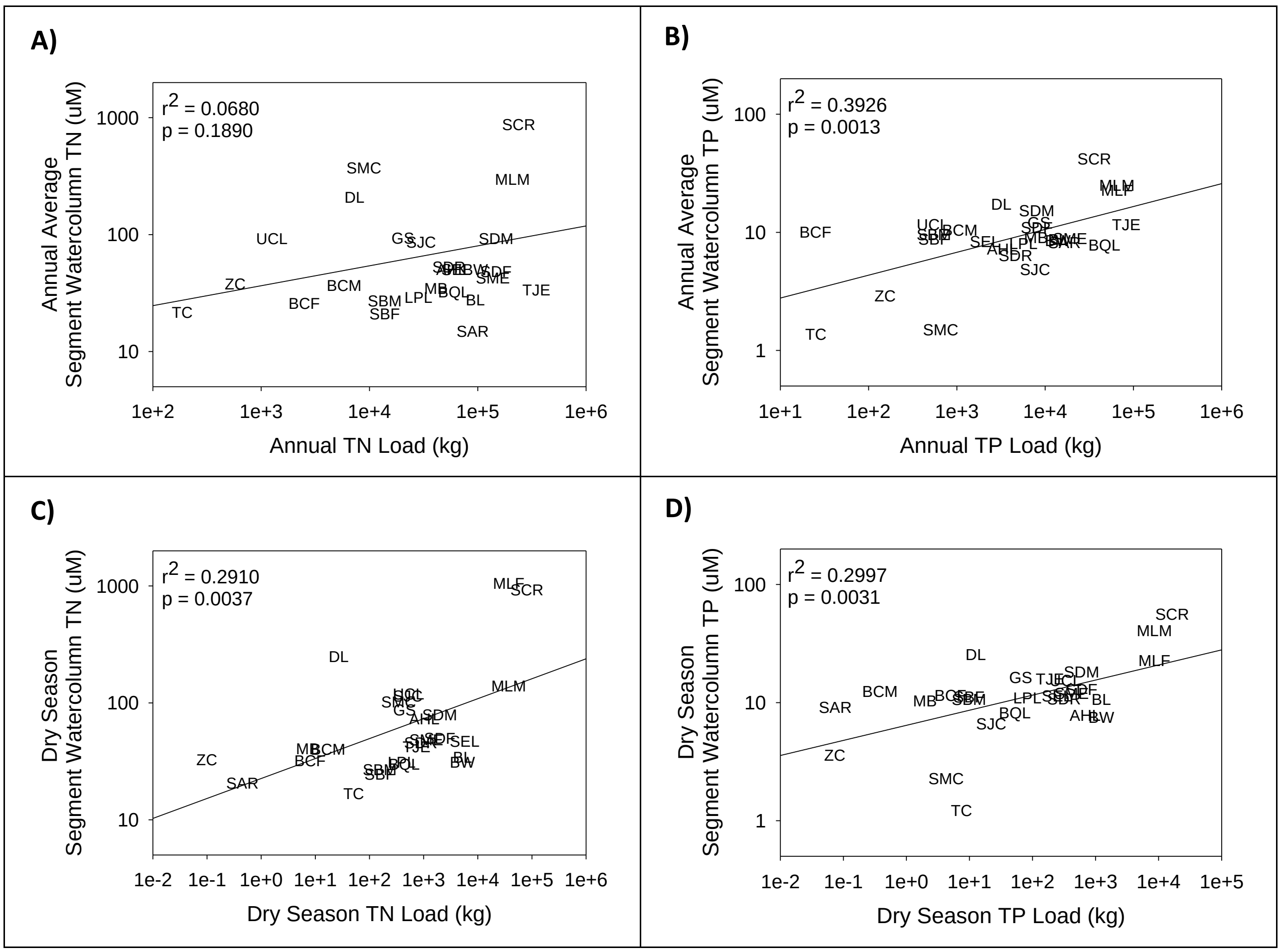

Figure V-3. Least-squares regression results for annual average water column TN and annual TN load (A), annual average water column TP and annual TP load (B), dry season average water column TN and dry season TN load (C), and dry season average water column TP and dry season TP load (D).

Relationship Between Water Column Nutrient Concentrations and Aquatic Primary Producer Biomass

Both macroalgae and phytoplankton biomass showed significant relationships with nutrient concentrations, with the strength of the least-squares regression relationship depending on temporal scale in which the data were averaged (annual average, wet season/winter or dry season/summer) (Table V-2; Figure V-4).

Macroalgal biomass was significantly, positively correlated with TN, PN, DON, TP and PP; the least-squares fit was greatest with annually-averaged biomass and nutrient concentration data (R^2 = 0.15 for TP and R^2 = 0.32 for TN; Table V-3). Of the nutrient species, dry season and peak season macroalgal biomass (when macroalgae biomass was highest in each segment) was most strongly related to DON. There was no significant relationship between dissolved inorganic nutrients (DIN or ortho-PO_4) and macroalgal abundance.

Table V-2. Results of least-squares regressions between estuarine water column nutrient concentrations and macroalgae and phytoplankton biomass (SCR excluded from analysis macroalgal analysis).

Aquatic Primary Producer	Nutrient Species	Annual Average[1]		Wet Season Average[2]		Dry Season Average[3]		Peak Season[4]	
		R^2	p-value	R^2	p-value	R^2	p-value	R^2	p-value
Macroalgae	TN	**0.3200**	**0.0026**	0.0083	0.6577	**0.2936**	**0.0043**	**0.2545**	**0.0078**
	PN	**0.2253**	**0.0143**	0.0000	0.9967	**0.2589**	**0.0078**	**0.1832**	**0.0291**
	DON	**0.4015**	**0.0005**	0.0051	0.7278	**0.3068**	**0.0033**	**0.2239**	**0.0146**
	DIN	0.0532	0.1529	0.0283	0.4112	0.0133	0.5755	0.0551	0.2482
	TP	**0.1589**	**0.0437**	0.0485	0.2796	0.1500	0.0506	0.0792	0.1636
	PP	**0.2327**	**0.0126**	0.0526	0.2598	**0.2044**	**0.0204**	**0.1752**	**0.0333**
	DOP	0.0555	0.2467	0.1045	0.1072	0.0274	0.4189	0.0127	0.5842
	PO_4	0.0716	0.1864	0.0043	0.7508	0.1295	0.0709	0.0495	0.2747
Phytoplankton	TN	**0.4405**	**0.0002**	**0.3617**	**0.0009**	**0.4707**	**<0.0001**	**0.4924**	**<0.0001**
	PN	**0.3513**	**0.0011**	**0.2930**	**0.0036**	**0.4478**	**0.0001**	**0.4352**	**0.0002**
	DON	**0.5395**	**<0.0001**	**0.4304**	**0.0002**	**0.3396**	**0.0014**	**0.3779**	**0.0060**
	DIN	**0.2050**	**0.0177**	**0.1760**	**0.0294**	**0.1660**	**0.0344**	**0.2143**	**0.0150**
	TP	0.0986	0.1107	0.0455	0.2855	0.0505	0.2596	0.0624	0.2087
	PP	**0.1959**	**0.0208**	**0.1780**	**0.0284**	0.0013	0.8602	0.0339	0.3579
	POP	0.0136	0.5617	0.0072	0.6736	0.0097	0.6295	0.0004	0.9218
	PO_4	0.0643	0.2019	0.0001	0.9565	0.0932	0.1214	0.0023	0.8114

[1] Annual Average - average biomass for all six sampling periods
[2] Wet Season Average- average biomass for sampling periods 1, 2 and 3 (November, January and March)
[3] Dry Season Average- average biomass for sampling periods 4, 5, and 6 (May, July, September)
[4] Peak Season- macroalgae = biomass and concentrations averaged from the two consecutive periods of highest abundance; phytoplankton = 90th percentile of annual CHLa data, and nutrient concentration during bloom

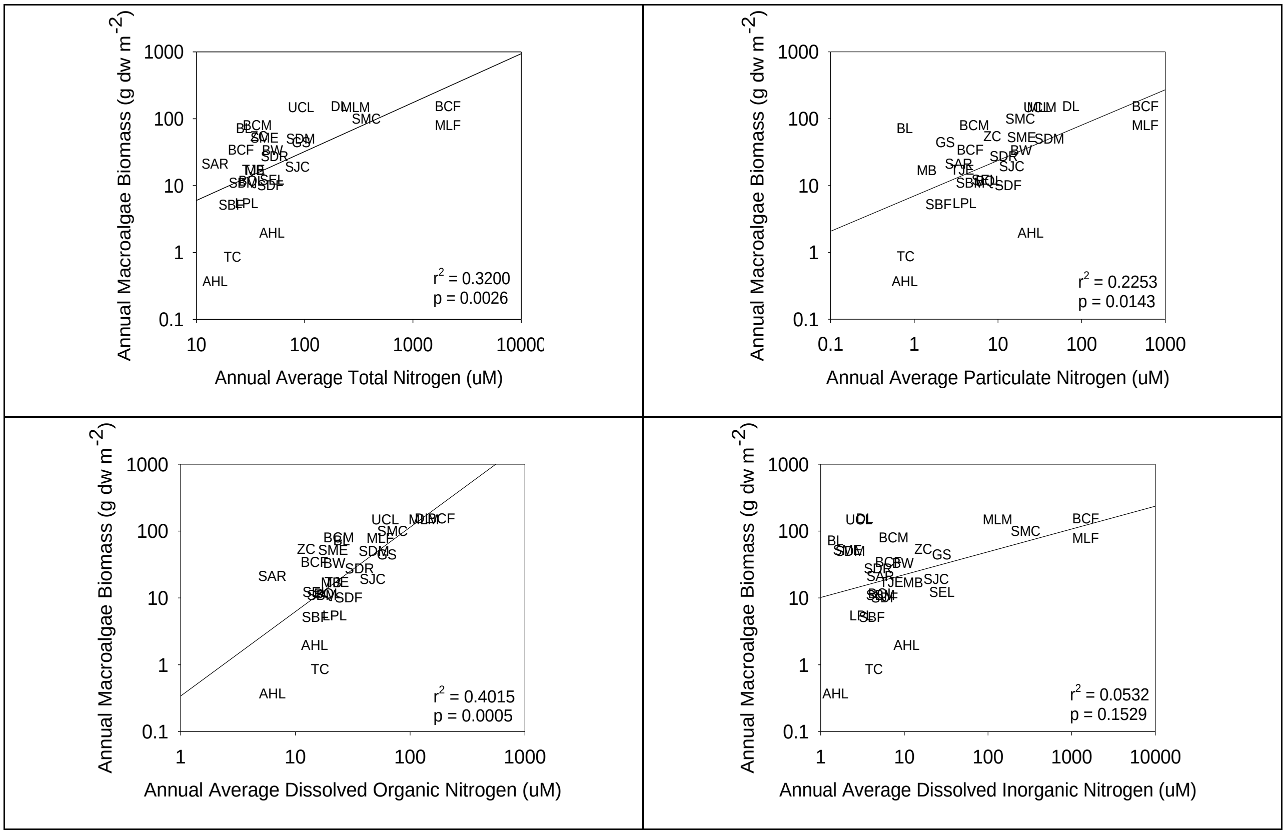

Figure V-4. Relationship between annual average macroalgae biomass and nitrogen species in water column at the segment site.

Table V-3. Regression equations for strongest least-squares regression relationships between primary producer response and estuarine water column nutrient concentrations.

Y	X	Regression Equation
Annual Average Macroalgae Biomass	Annual Avg TN	$\log(Anual\ Avg\ Macroalgae\ Biomass) = [0.73 * \log(TN)] + 0.05$
	Annual Avg PN	$\log(Anual\ Avg\ Macroalgae\ Biomass) = [0.53 * \log(TN)] + 0.85$
	Annual Avg DON	$\log(Anual\ Avg\ Macroalgae\ Biomass) = [1.26 * \log(TN)] - 0.47$
	Annual Avg DIN	$\log(Anual\ Avg\ Macroalgae\ Biomass) = [0.34 * \log(TN)] + 1.01$
	Annual Avg TP	$\log(Anual\ Avg\ Macroalgae\ Biomass) = [1.13 * \log(TN)] + 0.33$
	Annual Avg PP	$\log(Anual\ Avg\ Macroalgae\ Biomass) = [0.32 * \log(TN)] + 1.48$
	Annual Avg DOP	$\log(Anual\ Avg\ Macroalgae\ Biomass) = [0.82 * \log(TN)] + 0.81$
	Annual Avg DIP	$\log(Anual\ Avg\ Macroalgae\ Biomass) = [0.62 * \log(TN)] + 1.27$
Summer Average Phytoplankton Biomass	Summer Avg TN	$\log(Summer\ CHLa) = [0.71 * \log(TN)] - 0.63$
	Summer Avg PN	$\log(Summer\ CHLa) = [0.41 * \log(TN)] + 0.23$
	Summer Avg DON	$\log(Summer\ CHLa) = [0.68 * \log(TN)] - 0.39$
	Summer Avg DIN	$\log(Summer\ CHLa) = [0.22 * \log(TN)] + 0.46$
	Summer Avg TP	$\log(Summer\ CHLa) = [0.25 * \log(TN)] + 0.38$
	Summer Avg PP	$\log(Summer\ CHLa) = [0.03 * \log(TN)] + 0.64$
	Summer Avg DOP	$\log(Summer\ CHLa) = [0.04 * \log(TN)] + 0.60$
	Summer Avg DIP	$\log(Summer\ CHLa) = [0.16 * \log(TN)] + 0.60$

The strength of the regression relationships between water column nutrient concentrations and macroalgal biomass appear to be driven by the extreme ends of the disturbance gradient (Figure V-4). Segments span a range from high nutrients and high biomass (MLF,SMC), to sites with low nutrients and low biomass (TC, SBF, BQL). On a segment by segment basis, when macroalgae biomass is high in the segment, water column TN is low, particularly the dissolved inorganic fraction (Figure V-5).

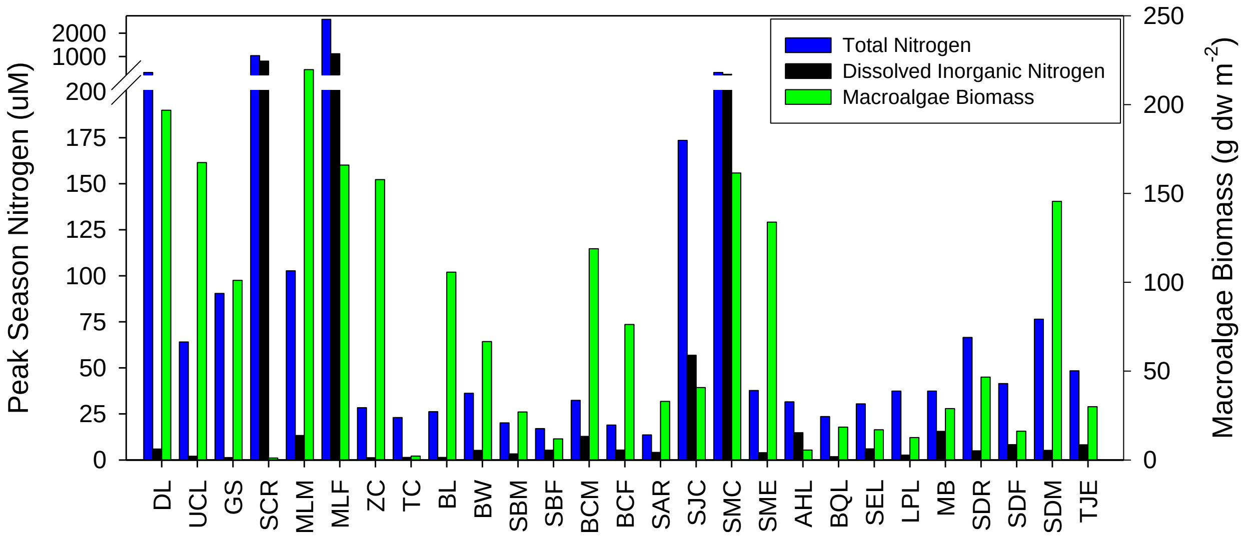

Figure V-5. Water column nitrogen concentration during period of peak macroalgae and phytoplankton biomass.

Phytoplankton biomass had a significant, positive correlation with TN, PN, DON, DIN, and PP; the least-squares fit was greatest for peak season (90th percentile of annual data) and TN concentration data (R^2 = 0.06 for TP and R^2 = 0.49 for TN; Table V-2, Figure V-6), but was significant for all nitrogen species during averaging periods considered as well. During the wet season, the strongest relationship was between phytoplankton biomass and DON (R^2 = 0.43). During the dry and peak seasons the relationship was strongest between phytoplankton biomass and PN (R^2 = 0.45) (Table V-3).

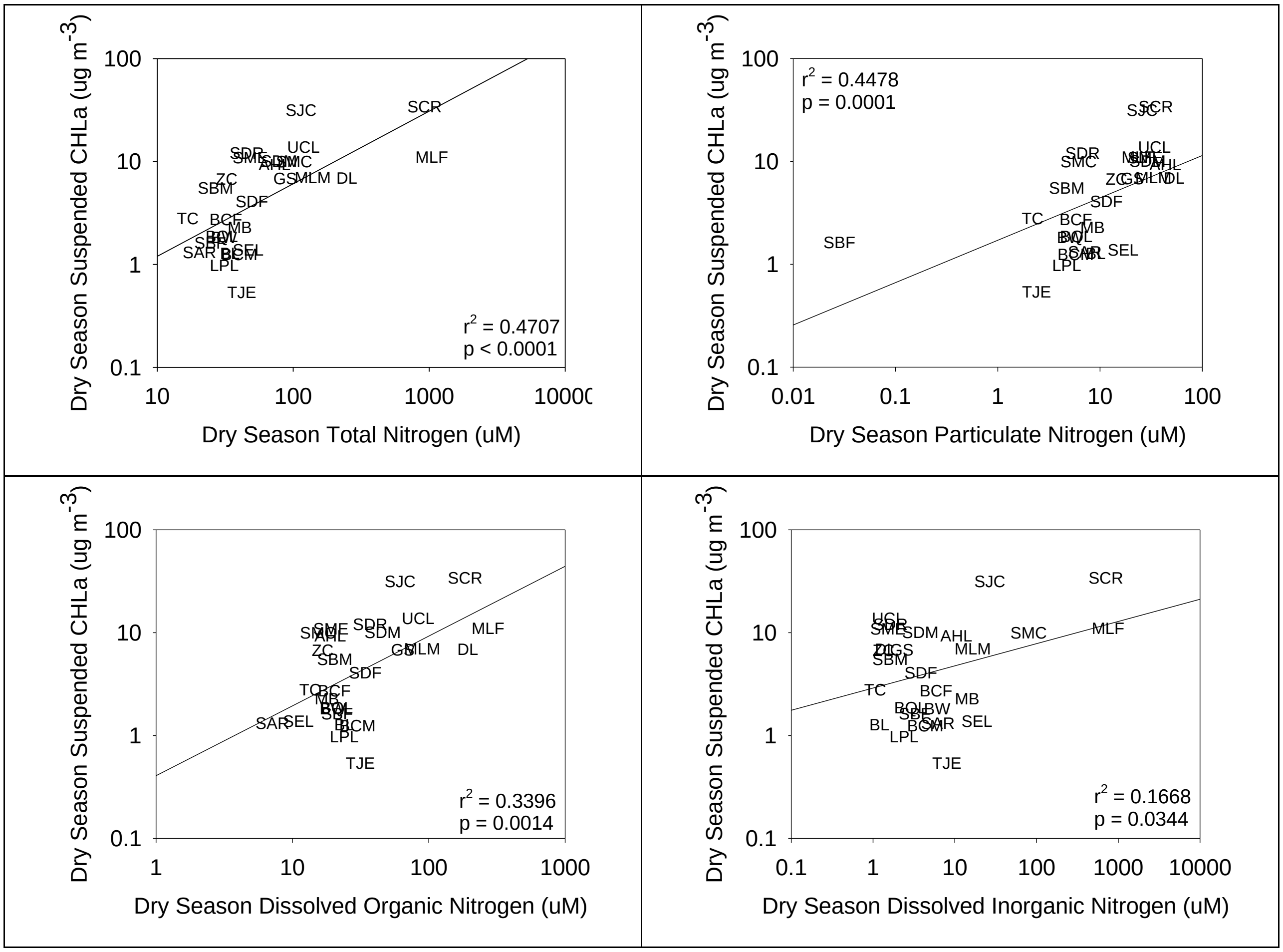

Figure V-6. Least-squares regression relationship between dry season averaged phytoplankton biomass for each segment and nutrient species in water column at the segment site.

Relationship Between Watershed Nutrient Loads and Aquatic Primary Producer Response

Macroalgae and phytoplankton biomass had significant, positive relationships with TN and TP loads; these relationships were improved when the volume of the estuary and residence time of water in the estuary are taken into account (Table V-4, Figures V-7 and V-8). For macroalgae, volume- and residence time-normalized annual TN and TP loads and wet weather TN and TP loads were significant (with R^2 ranging from 0.15 to 0.26 for TN and 0.16 to 0.33 for TP (Table V-4). For phytoplankton, only dry season TN and TP loads were significantly related to suspended chlorophyll *a* (R^2 ranging from 0.16 to 0.28 for TN and 0.16 to 0.32 for TP; Table V-4). Furthermore, the period over which the biomass data was integrated also had an effect on the strength of the relationship between nutrient loads and primary producer response. For macroalgae, while the relationship between nutrient loads and biomass was significant for all data integration periods, the strength of the relationship was greatest when the dry season average biomass was used as the response indicator (Table V-5). For phytoplankton, the load-response relationship was stronger for annual averaged chlorophyll *a* compared to peak chlorophyll *a* (90th percentile) (Table V-5).

Table V-4. Least-squares regression result for nutrient load- aquatic primary producer response scenarios.

Load	Normalization	Macroalgae Biomass								Phytoplankton Biomass			
		Maximum Period		Peak Season		Dry Season		Annual Average		Annual Average Suspended CHL a		90%tile Suspended CHL a	
		R^2	p-value	R^2	p-value	R^2	p-value	R^2	p-value	R^2	p-value	R^2	p-value
Annual TN	None	0.0019	0.8327	0.0051	0.7282	0.0147	0.5555	0.0139	0.5661	0.0076	0.6650	0.0016	0.8420
	Residence Time	0.0851	0.1481	0.0877	0.1419	0.1113	0.0958	0.1071	0.1027	0.1076	0.0948	0.0856	0.1385
	Volume	0.0445	0.2994	0.0533	0.2565	0.1284	0.0722	0.0496	0.2743	0.0019	0.8271	0.0003	0.9363
	Volume and Residence Time	**0.1526**	**0.0484**	**0.1535**	**0.0478**	**0.2505**	**0.0092**	**0.1490**	**0.0500**	0.0290	0.3959	0.0393	0.3213
Dry Season TN	None	0.0005	0.9165	0.0029	0.7926	0.0247	0.4432	0.0103	0.6213	**0.1607**	**0.0382**	0.1206	0.0759
	Residence Time	0.0362	0.3520	0.0430	0.3096	0.0899	0.1366	0.0603	0.2267	**0.2831**	**0.0043**	**0.2360**	**0.0102**
	Volume	0.0189	0.5031	0.0266	0.4258	0.0960	0.1235	0.0301	0.3969	0.0749	0.1673	0.0728	0.1735
	Volume and Residence Time	0.0316	0.3853	0.0407	0.3229	0.0985	0.1185	0.0387	0.3357	**0.1812**	**0.0269**	**0.1825**	**0.0263**
Wet Weather TN	None	0.0048	0.7380	0.0095	0.6354	0.0130	0.5787	0.0188	0.5044	0.0003	0.9275	0.0004	0.9245
	Residence Time	0.0849	0.1486	0.0892	0.1384	0.0951	0.1254	0.1058	0.1050	0.0643	0.2017	0.0507	0.2589
	Volume	0.0590	0.2319	0.0706	0.1894	0.1378	0.0619	0.0654	0.2073	0.0100	0.6189	0.0045	0.7407
	Volume and Residence Time	**0.1748**	**0.0335**	**0.1782**	**0.0317**	**0.2603**	**0.0078**	**0.1648**	**0.0396**	0.0160	0.5300	0.0254	0.4275
Annual TP	None	0.0032	0.3792	0.0374	0.3435	0.1115	0.0954	0.0552	0.2479	0.0138	0.5600	0.0094	0.6313
	Residence Time	0.1463	0.0538	0.1419	0.0578	**0.2454**	**0.0101**	**0.1641**	**0.0400**	0.1071	0.0956	0.0983	0.1113
	Volume	0.0786	0.1654	0.0831	0.1533	**0.2170**	**0.0165**	0.0797	0.1624	0.0000	0.9774	0.0011	0.8704
	Volume and Residence Time	**0.1822**	**0.0297**	**0.1768**	**0.0325**	**0.3343**	**0.0020**	**0.1671**	**0.0381**	0.0298	0.3891	0.0443	0.2918
Dry Season TP	None	0.0028	0.7984	0.0072	0.6798	0.0275	0.4180	0.0118	0.5973	**0.1950**	**0.0211**	**0.1593**	**0.0392**
	Residence Time	0.0462	0.2917	0.0539	0.2536	0.0928	0.1303	0.0617	0.2210	**0.3212**	**0.0021**	**0.2815**	**0.0044**
	Volume	0.0278	0.4160	0.0373	0.3443	0.1053	0.1058	0.0338	0.3687	0.1039	0.1010	0.1071	0.0956
	Volume and Residence Time	0.0407	0.3229	0.0512	0.2663	0.1037	0.1086	0.0413	0.3196	**0.2156**	**0.0147**	**0.2245**	**0.0125**
Wet Weather TP	None	0.0356	0.3558	0.0404	0.3249	0.1124	0.0941	0.0577	0.2373	0.0061	0.6978	0.0039	0.7565
	Residence Time	0.1460	0.0541	0.1413	0.0584	**0.2397**	**0.0111**	**0.1621**	**0.0414**	0.0838	0.1431	0.0782	0.1577
	Volume	0.0590	0.2319	0.0706	0.1894	0.1378	0.0619	0.0654	0.2073	0.0100	0.6189	0.0045	0.7407
	Volume and Residence Time	**0.1744**	**0.0335**	**0.1782**	**0.0317**	**0.2603**	**0.0078**	**0.1648**	**0.0396**	0.0160	0.5300	0.0254	0.4275

Table V-5. Regression equations for strongest least-squares regression relationships between primary producer response and watershed nutrient loading.

Y	X	Regression Equation
Peak Season Macroalgae Biomass	Volume and Residence Time Normalized Annual TN Load	$\log(Peak\ Macroalgae\ Biomass) = [0.15 * \log(\frac{Annual\ TN\ load}{365\ days} * res.time * \frac{1}{Estuary\ Vol})] + 1.7$
	Volume and Residence Time Normalized Annual TP Load	$\log(Peak\ Macroalgae\ Biomass) = [0.14 * \log(\frac{Annual\ TP\ load}{365\ days} * res.time * \frac{1}{Estuary\ Vol})] + 1.8$
Summer Macroalgae Biomass	Volume and Residence Time Normalized Annual TN Load	$\log(Summer\ Macroalgae\ Biomass) = [0.25 * \log(\frac{Annual\ TN\ load}{365\ days} * res.time * \frac{1}{Estuary\ Vol})] + 1.4$
	Volume and Residence Time Normalized Annual TP Load	$\log(Summer\ Macroalgae\ Biomass) = [0.24 * \log(\frac{Annual\ TP\ load}{365\ days} * res.time * \frac{1}{Estuary\ Vol})] + 1.6$
Annual Average Phytoplankton Biomass	Volume and Residence Time Normalized Dry Season TN Load	$\log(Annual\ Avg\ CHLa) = [0.10 * \log(\frac{Annual\ TP\ load}{365\ days} * res.time * \frac{1}{Estuary\ Vol})] + 0.82$
	Volume and Residence Time Normalized Dry Season TP Load	$\log(Annual\ Avg\ CHLa) = [0.12 * \log(\frac{Annual\ TP\ load}{365\ days} * res.time * \frac{1}{Estuary\ Vol})] + 0.93$

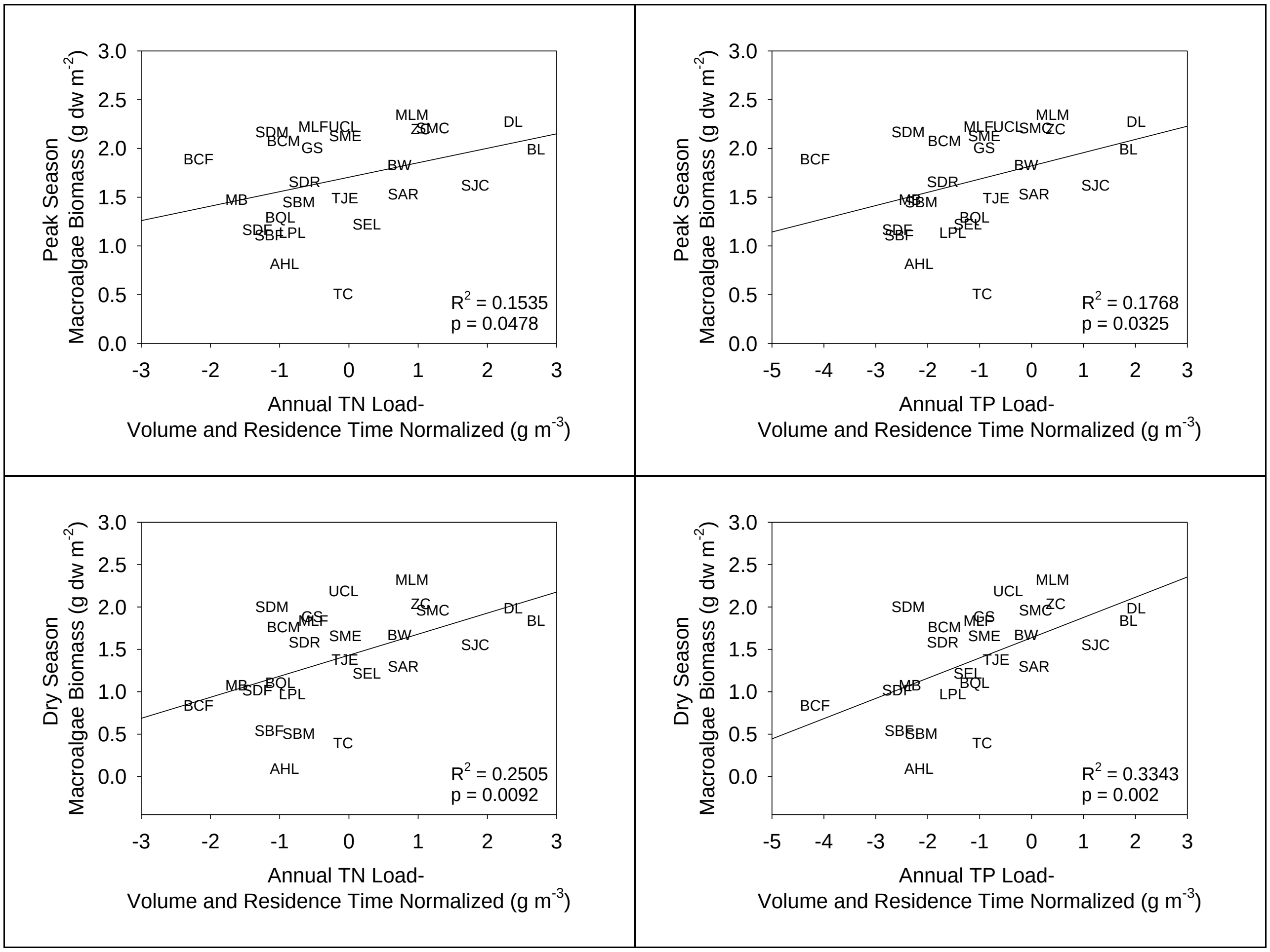

Figure V-7. Relationships between watershed nutrient loads and macroalgae response.

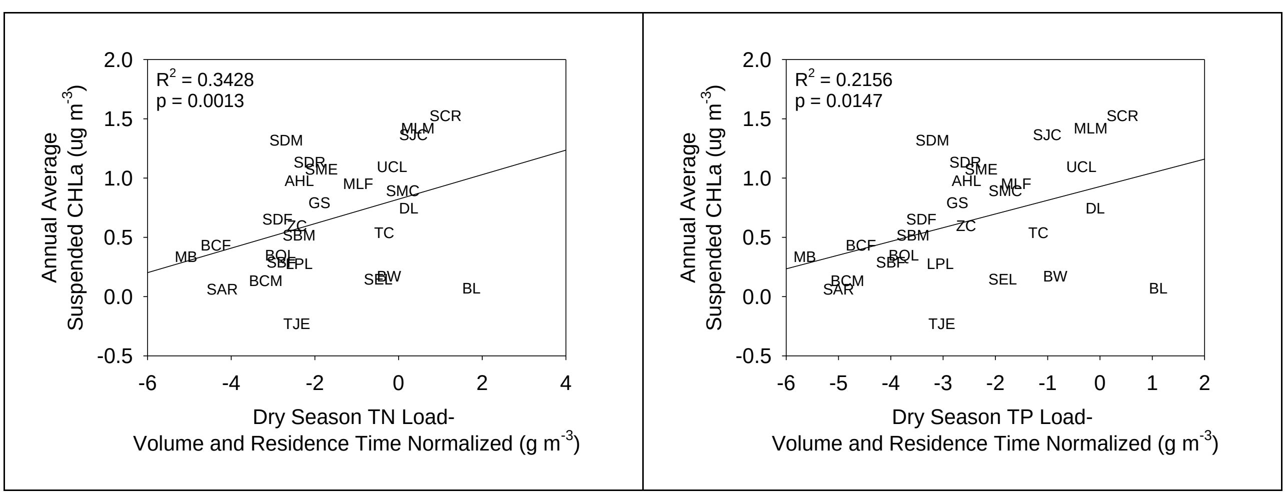

Figure V-8. Relationships between watershed nutrient loads and phytoplankton response.

Discussion

A number of studies have found linkages between algal response indicators and either loads or concentrations for both macroalgae and phytoplankton (Valiela *et al.* 1990, Conley *et al.* 2000, Pinckney *et al.* 2001, Smith 2006, Boynton and Kemp 2008, Madden *et al.* 2010). In this study, we found that both macroalgal and phytoplankton biomass had a significant correlation with estuarine water column nutrients concentrations as well as watershed nutrient loads. Important points emerge from the analyses: 1) the relationship of nutrient inputs (watershed nutrient loads and estuarine watercolumn concentrations) and aquatic primary producer biomass was significant but lacked precision, and was better for phytoplankton than macroalgae; 2) estuarine water column concentrations generally had a better correlation with aquatic primary producer biomass than nutrient loads, though loads and concentrations were correlated on some timescales; 3) selecting the appropriate timescales over which to average the data is important to the outcome of the analysis; and 4) total nutrients were better correlated with biomass than dissolved inorganic nutrients.

In general, variations in N loading rates are reflected in concentrations of N in receiving water bodies, particularly when residence time of that water body is long (on the order of weeks (Conley *et al.* 2000, Smith *et al.* 2005, Boynton and Kemp 2008, Hejzlar *et al.* 2009). In this study, we found that dry season riverine TN and TP loads, when residence time was the longest, explained 20-30% of the variation in estuarine water column TN and TP concentrations. Mean TN concentrations were significantly correlated to TN loading for five sub-systems of Chesapeake Bay averaged over a decadal period (Boynton *et al.* 2008). Conley *et al*. (Conley *et al.* 2000) reported that on an annual basis about 70% on the variation in TN concentration could be explained by variation in TN loads in 81 Danish estuaries by dominated agricultural inputs.

In this study, nutrient concentrations generally explained a greater percentage of variation in phytoplankton biomass than macroalgal biomass. The bulk of literature citations linking aquatic primary producer biomass to nutrient concentrations or loads are for phytoplankton biomass ((Boynton *et al.* 2008), Table V-6, (Smith 2006, Madden *et al.* 2010); Table V-6). PN and TN accounted for 43-47% of the variation in dry season phytoplankton biomass; this finding can be explained by the fact that measurement of particulate nutrients or total nutrients includes phytoplankton biomass. In contrast, several studies cite the lack of relationship between nutrient concentrations and macroalgae (Valiela *et al.* 1997, Kennison *et al.* 2003, McGlathery *et al.* 2007). More studies link macroalgae to loads (Valiela *et al.* 1992, Valiela *et al.* 1997, Hauxwell *et al.* 1998, Conley *et al.* 2000, Fox *et al.* 2008). Macroalgae have very high nutrient uptake rates and can rapidly take up large pulses of inorganic nitrogen (Fujita 1985, Pedersen and Borum 1997, Lotze and Schramm 2000, Runcie *et al.* 2003). These physiological adaptations often result in high abundances of macroalgae co-occurring with low or non-detectable concentrations water column concentrations, particularly during bloom initiation, as noted in some segments in this study. During latter stages of blooms, particularly during senescence, macroalgae are known to release large amounts of dissolved organic matter (Anderson and Zeutschel 1970, Valiela *et al.* 1997, Pregnall 19893) because they fix more carbon and nutrients than they require and exude the unused dissolved organic matter, releasing up to 39% of their gross production during bloom periods

(Velimirov 1986). SCB segments were characterized by high concentrations of DON, the form of N which explained the most variation in macroalgal biomass.

Table V-6. Modeled relationships between nutrient loading and phytoplankton response in world estuaries. (From Boynton and Kemp 2008).

Location	Variable, X (units)	Variable, Y	r^2 / n	Reference
Multiple estuaries	TN-loading (g N m^{-2} y^{-1})	Phytoplankton Production	0.60 / 14	Boynton et al. 1982
SF Bay and other estuaries	Composite parameter X = f(B, Z_p, I_0)		0.82 / 211	Cole and Cloern 1987
Narragansett Bay	Composite parameter X = f(B, Z_p, I_0)		0.62 / 1010	Keller 1988
Multiple estuaries	DIN-loading (mol N m^{-2} y^{-1})		0.93 / 19	Nixon et al. 1996
Multiple estuaries	TN-loading (g N m^{-2} y^{-1})		0.36 / 51	Borum and Sand-Jensen 1996
Boston Harbor	Composite parameter X = f(B, Z_p, I_0)		0.66 / 12	Kelly and Doering 1997
Waquoit Bay system	Annual average DIN conc (μM)		0.61 / 12	Valiela et al. 2001
Chesapeake Bay	TN(x_1), TP(x_2) load (kg mo^{-1})		0.67 / 11	Harding et al. 2002
Multiple estuaries	DIN (mM m^{-3}); tidal range (m)	Phytoplankton Biomass	na / 163	Monbet 1992
Multiple systems / MERL	DIN input (mmol m^{-3} y^{-1})		na / 34	Nixon 1992
Ches Bay mesohaline	River flow (m^3 d^{-1}); proxy for N-load		0.70 / 34	Harding et al. 1992
Maryland lagoons	TN load (g N m^{-2} y^{-1})		0.96 / 9	Boynton et al. 1996
Danish coastal waters	TN concentration (ug l^{-1})		0.64 / 168	Borum 1996
Canadian estuaries	TN concentration (ug l^{-1})		0.72 / 15	Meeuwig 1999
Ches Bay and Tributaries	TN Load; (mg N m^{-2} yr^{-1}) (R_{time}, yrs)$^{-1}$		0.82 / 17	Boynton and Kemp 2000
Danish estuaries	TN concentration (ug N l^{-1})		0.30 / 1347	Nielsen et al. 2002

With either macroalgae or phytoplankton biomass, dissolved inorganic nutrients explained the least amount of variation. Similar studies have documented this. Smith (Smith 2006) documented that relationship between nutrient concentrations and phytoplankton biomass was generally stronger with TN than DIN. The concentration of a dissolved inorganic nutrient measurable in the water column represents the instantaneous net “remainder” after processing by all other factors. Thus TN or TP concentrations are a better measure of integrated exposure over time than nutrient availability than dissolved inorganic N or P.

Averaging period was important, both in terms of load or concentration data as well as aquatic primary producer biomass. Other studies report success in relating phytoplankton to both external nutrient loads and in-situ nutrient concentrations in estuaries, particularly when data are averaged over annual or decadal time periods (Boynton *et al.* 2008, Boynton and Kemp 2008). In this study, annually-averaged was generally the best approach, with some exceptions. Averaged annual estuarine nutrient concentrations had the highest correlation with annually averaged macroalgal biomass, followed by dry season and peak season biomass. For loads, annually- and wet season-averaged TN loads had the highest correlation with macroalgal biomass, but averaging of macroalgal data had not real effect on strength of correlation. For phytoplankton, all methods of averaging nutrient concentrations were significant, while averaging of phytoplankton performed best with peak season (90th percentile), with dry season and annual concentrations less predictable.

There has been a great deal of discussion among scientists and managers about whether estuarine surface water nutrient concentrations or nutrient loading to the estuary is a more relevant as target for management of eutrophication. Estuarine nutrient concentrations were equal to or slightly better than riverine nutrient loads in predicting aquatic primary producer biomass, though overall the precision of both types of least-squares regressions were poor. There are several reasons for this. First, riverine nutrient loads represent only a partial accounting of total sources of nutrients available for aquatic primary producers; for example, benthic fluxes of nutrients in particularly in shallow lagoonal estuaries typical of SCB estuaries have been found to account for more than 90% of total nutrient loads (Sutula *et al.* 2004, Sutula *et al.* 2006). Second, estimates of residence time and volume were significant co-factors that served to increase the statistical significance of the load-response relationship, similar to findings of Dettman *et al*. (Dettmann 2001) and Nixon *et al*. (Nixon and al. 1996). We utilized existing data sources to constrain these numbers, yet acknowledge the tremendous uncertainty in these estimates. We recommend improving them through the development of, at minimum, comprehensive surveys of estuarine merged bathymetry-topography and the development of one dimensional box models better constrain estuarine hydrology. Third, additional site-specific factors including bathymetry, and light limitation due to suspended sediment load are also known to have a strong effect on the response of primary producer communities to nutrient loading and are unaccounted for in the statistical models described above (Painting *et al.* 2007a, Painting *et al.* 2007b). Finally, the data set was of limited sample size, so the strength of the least-squares regression for loads or nutrient concentration and aquatic primary producer biomass seemed to be driven by the extremes in the data set (very high loads or concentrations and biomass, versus very low concentrations and biomass). Consequently these relationships seem more indicative of a disturbance gradient in nutrient loading rather than a predictive model of ecosystem response to nutrient loading. While the positive relationships between estuarine condition and nutrient loads gives confidence in the use of ambient nutrient concentrations and primary producer biomass as indicators of eutrophication in SCB estuaries, these models lack the precision to set site specific water quality goals to improve ecosystem health.

In contrast to algae, extent of low DO events had no significant correlation with N and P loads; instead, it was strongly related to sediment organic matter (OM) content (Section IV). Macroalgae was also significantly correlated with sediment OM. Sediment OM generally increases along a gradient of increasing eutrophication, with increased amounts of reflecting accumulation of external loading and/or within-estuary production and accumulation of algae over decadal time-scales. Thus, DO and algae indicators integrate the effects of increased nutrient loading over very different time-scales. aquatic primary producer biomass reflects a more immediate response to nutrient loads entering on that particular year, while low DO is largely driven by the combination of OM loading and aquatic primary producer biomass which has accumulated over time. These findings have important implications for how different response indicators can be used for management of eutrophication in estuaries. If nutrient loads are reduced, one may expect to see a response in algal blooms relatively quickly, while the time required to address hypoxia problems in an estuary greatly extended.

In conclusion, several studies have demonstrated the shortcomings of using estuarine nutrient concentrations or loads alone to predict ecosystem response to eutrophication (Cloern 2001, Dettmann 2001, Kennison *et al.* 2003). Estuaries are highly variable in how they respond to nutrient loading due to differences in physiographic setting, salinity regime, frequency and timing of freshwater flows, magnitude of tidal forcing, sediment load, stratification, residence time, denitrification, etc. (Pinckney *et al.* 2001, Zaldivar *et al.* 2008). This combination of "co-factors" results in differences in the dominant primary producer communities (i.e., phytoplankton, macroalgae, benthic algae, submerged aquatic vegetation, emergent macrophytes) and creates variability in the pathways that control how nutrients cycle within the estuary. At times, these co-factors can play a larger role in mitigating estuarine response to nutrient loads or estuarine water column concentrations, blurring or completely obscuring simple nutrient limitation of primary production (Cloern 2010). To improve load-response models for SCB estuaries we recommend several actions. First, the appropriate timescales over which the data should be averaged must be determined for each estuary. SCB estuaries have peak primary production at different times during the year (SectionIII), averaging loads over the period critical for primary production could improve model output (e.g., systems that have peak production in winter may be more closely related to wet weather loads and systems that have peak production in summer may be more closely related to dry season loads). Second, a more complete accounting of loads into the systems may greatly improve predictability. Benthic flux studies in a handful of SCB estuaries have shown that the sediments may act as a source or sink of nutrients depending on the system (Sutula *et al.* 2004, Sutula *et al.* 2006), thus having a full accounting of nutrient sources and sinks should improve model output. Finally, incorporation of "co-factors" (bathymetry, suspended solids, presence of fringing marshes etc.) that are known to affect ecological response to nutrient enrichment should also improve model output.

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VI. ASSESSMENT OF EUTROPHICATION IN ESTUARIES OF THE SOUTHERN CALIFORNIA BIGHT: A SYNTHESIS OF FINDINGS

Introduction and Major Study Questions

The estuaries of the southern California, found in a distinct region that extends from Point Conception to Punta Banda, Baja Mexico, are an important resource for biodiversity, support of commercial and recreational fisheries, migratory birds, endangered species, as well as tourism. These estuaries are at risk, due to habitat loss, fragmentation, and increased loading of contaminants from urbanized and agricultural watersheds. Nutrients are a major form of contaminant loading, particularly from point sources such as industrial and municipal effluent and non-point sources such runoff from agricultural and residential land uses and atmospheric deposition.

While nitrogen (N) and phosphorus (P) are required to support all life forms, too much causes problems. Nutrient pollution causes an over-growth of algae and aquatic plants, leading to reduced dissolved oxygen (DO) concentrations, reduced biodiversity and changes in food webs. This collection of symptoms is referred to as "eutrophication." Eutrophication is recognized as one of the leading impairments of water quality in the United States, yet, despite the large number of estuaries in the Southern California Bight (SCB), little data are available on extent of eutrophication and the relationship with watershed nutrient loads. Only three of the SCB's 76 estuaries had sufficient data to be included in the 2007 National Oceanographic and Atmospheric Administration (NOAA) National Estuary Eutrophication Assessment. As the State Water Resources Control Board (SWRCB) prepares to develop estuarine nutrient objectives, a heightened need exists to identify appropriate indicators, standard protocols and methods to interpret data, and establish linkages between nutrient inputs and symptoms of eutrophication to support improved nutrient management.

The Bight 2008 Estuarine Eutrophication Assessment provided an opportunity to conduct the first large scale assessment of estuarine eutrophication in southern California. In addition, the process could be used to inform the development of estuarine nutrient objectives and get early agreement on indicators and standard protocols. Working together, environmental managers from twenty-one organizations, including stormwater agencies, municipalities, state and federal regulatory agencies, and scientists joined forces to answer three basic questions:

1. What is the extent and magnitude of eutrophication in SCB estuaries?
2. Is there a difference in eutrophication between different classes of estuaries or by the degree of tidal flushing?
3. Is there a relationship between the symptoms of eutrophication and nutrient inputs?

Study Approach, Design, and Framework Used to Interpret Data. Magnitude of eutrophication was assessed using macroalgal abundance, phytoplankton biomass, microphytobenthos biomass, submerged aquatic vegetation biomass, and dissolved oxygen (DO). These indicators have scientifically well-vetted linkages to the ecosystem functions and beneficial uses of estuaries. Total N and P loading from the watershed, as well as 19 other physical and chemical parameters (including sediment and water column nutrient content, sediment grain-size, and continuously measured water column temperature, salinity, pH, turbidity, and water level) were also measured to determine how site-specific factors affect magnitude and extent of eutrophication.

Because eutrophication is highly spatially variable within an estuary, and because this was a regional assessment, we chose to report on targeted index area (segment) within many estuaries to get a broad estimate of extent of eutrophication across the region. A total of 27 segments in 23 estuaries were randomly selected from a comprehensive list of 76 estuaries. For the majority of systems, the segment represents 50 - 100 % of the estuarine area, but for a subset of systems (7 estuaries) the segment represents less than 25% of the total area. Segments were proportionally selected from the list to be able to look at differences by estuarine classes (enclosed bay, lagoon, river mouth estuaries) and degree of tidal exchange with the ocean (a.k.a. tidal inlet status: open, diked, closed). Each segment was located in a region of the estuary that is likely to have a longer residence time in order to capture where the symptoms of eutrophication would most likely be expressed. DO and phytoplankton biomass was assessed continuously while macroalgal biomass and other parameters assessed every other month from November 2008-October 2009.

Reporting on the extent of eutrophication requires a framework to interpret data. Towards this end, DO, macroalgae, and phytoplankton assessment frameworks for European Union Water Framework Directive (EU-WFD) were applied to the monitoring data set to assess extent of eutrophication. Ecological condition in each segment was classified into one of five categories from very high (minimally disturbed conditions), high, moderate, low, to very low (severely degraded condition) for each indicator.

Study Findings

Question 1: What is the Extent and Magnitude of Eutrophication in Southern California Bight Estuaries?

According to the EU-WFD framework, the Bight'08 assessment found that eutrophication was pervasive in the SCB segments monitored. The EU-WFD requires management action if ecological condition is listed as "moderate" or worse (Zaldivar *et al.* 2008). In SCB estuaries, 78% of segments using macroalgal abundance, 39% using phytoplankton biomass, and 63% using dissolved oxygen were categorized in "moderate" ecological condition or below indicating that they are affected by eutrophication (Figure VI-1). EU-WFD applies a "one out, all out" approach in determining ecological status wherein the lowest score for any single indicator becomes the overall score the waterbody (Borja *et al.* 2004). Applying this standard to the Bight'08 assessment, all but one of the segments (96%; SBF scored high or better in all assessed categories) assessed would require management action. Applying a multiple lines of evidence approach to classify these segments, in which at least one of the two algal indicators (primary symptom) and dissolved oxygen (a secondary symptom) fall in a category of moderate or worse, would result in 53% of segments requiring management action. Thus, over half of systems surveyed would require some management action, according to the EU-WFD framework. These findings should be interpreted with caution because segments from larger estuaries were located within the part of the estuary where we would be most likely to find a problem, if it exists. Furthermore, dissolved oxygen was measured in bottom water where hypoxia would most likely develop (compared to surface waters). Consequently, these study results represent a conservative estimate of extent and magnitude of eutrophication in SCB estuaries regionally and is not meant to be an exhaustive analysis of any single estuary. However, this study is the first ever regional assessment of eutrophication in SCB estuaries and can be used as a starting point for future assessments and management actions.

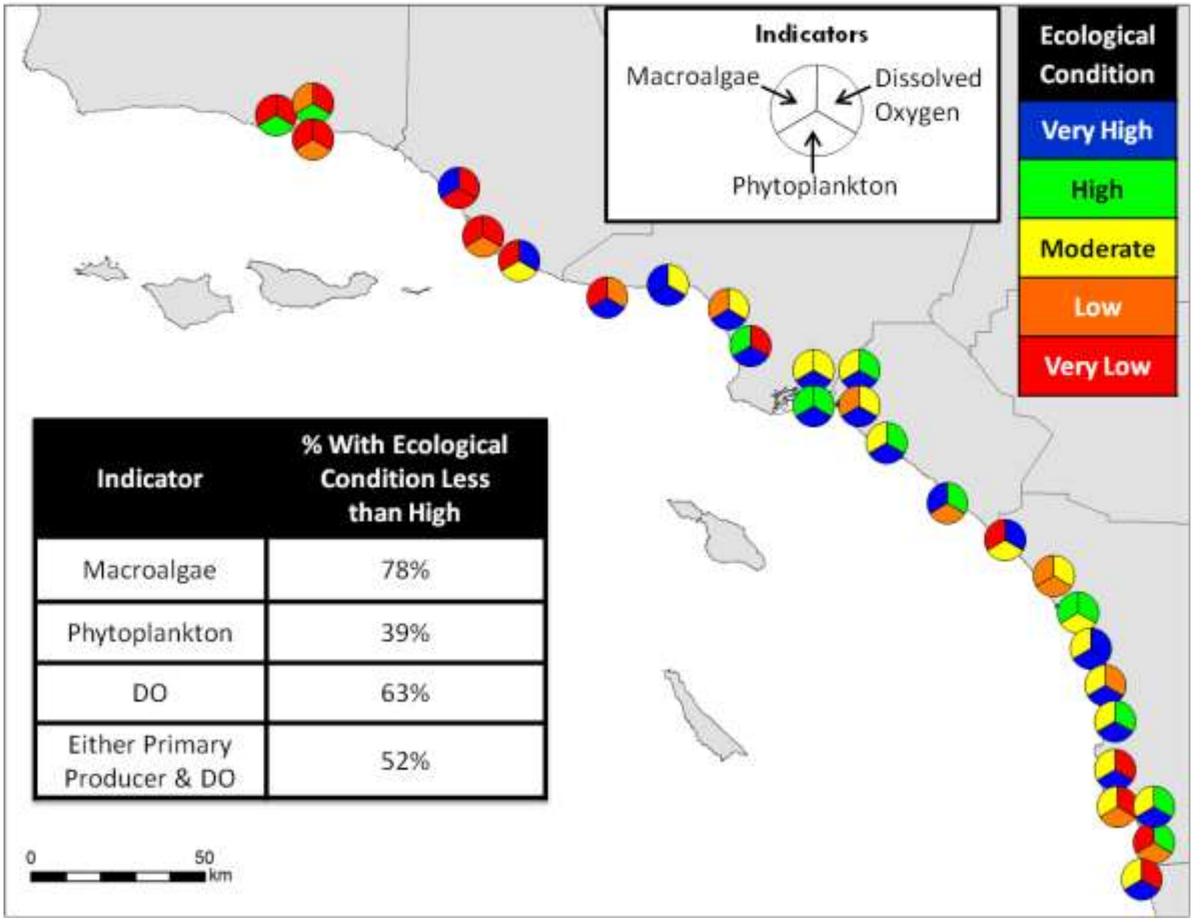

Figure VI-1. Ecological condition category determined for each estuarine segment using three response indicators of eutrophication: macroalgae, phytoplankton and dissolved oxygen concentration. Summary table shows the percent of segments falling below the "high" threshold for each indicator as well as a mulitimetric approach of one primary producer and dissolved oxygen.

The data can also be used to provide the relative ranks of the estuarine segments individually for each of the indicators, or overall by integrating across all indicators (Table II-7). Use of the data in this way provides context of the condition of the estuary, relative to other estuaries in our region. In addition to putting the segments into a regional context, this exercise helps to highlight which indicators are relevant in specific systems and how site specific factors can mediate the response of some indicators. The three response indicators (macroalgae, dissolved oxygen, and phytoplankton) did not necessarily score the segment the same (Table II-7). In some cases, this reflects difference in what algal group dominated (phytoplankton versus macroalgae). In other cases, the indicator is reflecting a different aspect of eutrophication (algae versus dissolved oxygen). Generally, macroalgae was the dominant primary producer, though several exceptions were found.

Question 2: Is There a Difference in Eutrophication between Different Classes of Estuaries or by the Degree of Tidal Flushing?

Environmental managers were interested testing the effect of estuarine class and degree of restricted tidal hydrology (i.e., inlet status) on extent of eutrophication. Enclosed bays are the largest, deepest estuaries, and have well-flushed, permanent connections with the ocean. In comparison, the smaller lagoons and river mouth estuaries, which have sand bars that form across their mouths, have intermittent restriction or complete closure of their tidal inlets. We hypothesized that extent of eutrophication would be higher in estuaries with more restricted hydrology (if all else is equal, lagoons would be more eutrophic than river mouths which are more eutrophic than enclosed bays; and/or, estuaries with a closed inlet are more eutrophic than estuaries with a restricted inlet, which are more eutrophic than estuaries with an open inlet) and that, within the same estuary, segments behind dikes or tide gates would be more eutrophic than those outside of such structures. Results of this study showed that estuarine class had no effect on extent of eutrophication and nutrient or organic matter loading was more important than inlet status in terms of nutrient impairment. However, macroalgal biomass decreased significantly as the tidally-induced variation in water level increased. In addition, the relationship between algal biomass and N and P loads became more significant when the volume and residence time of water of the estuary were both taken into account. Water residence time is largely driven by the status of the ocean inlet and volume is a function of the morphology of the estuary. Furthermore, extent of low DO and macroalgae biomass were significantly related to sediment organic matter, which is typically preserved in habitats that tend to deposit fine-grained sediment in areas of restricted hydrology. Among paired restricted and unrestricted ocean inlet segments, restricted segments were ranked lower for nutrient impairment compared to unrestricted segments in the same estuary. Thus, inlet status likely influences the extent of eutrophication, but is not necessarily more important than gradients in nutrient and organic matter loading, which can vary greatly on a site-specific level.

Question 3: Is Indicator Response Driven by Nutrient Loads or Nutrient Concentrations?

Eutrophication is assessed based on ecological response indicators, but impairment is managed largely by reducing nutrient inputs. This question sought to determine if relationships existed between nutrient inputs and the magnitude of eutrophication symptoms in SCB estuaries, and whether these relationships were stronger for estuarine nutrient concentrations or nutrient loads into the estuary. This question is important for two reasons. First, management strategies differ by whether the ultimate endpoint is nutrient loads (weight per unit time) into the estuary or estuarine water column nutrient concentrations. Strategies to control wet weather inputs, which provide most of the load, are different from strategies to reduce water column concentrations, which are more applicable during dry weather conditions. Second, the USEPA approach to nutrient objectives is driven by the assumption that estuarine nutrient concentrations are a good predictor of eutrophication. We used statistical models to determine the strength of the relationships between extent of eutrophication and nutrient concentrations or loads.

Watershed nutrient loads and estuarine water-column nutrient concentrations were both found to be significantly, positively correlated with aquatic primary producer biomass (i.e., macroalgae and phytoplankton). Several important points emerge from the analyses: 1) the relationship of nutrients and aquatic primary producer biomass was generally weak, though generally better for phytoplankton than macroalgae; 2) estuarine water column concentrations generally had a higher correlation with aquatic primary producer biomass than nutrient loads; 3) selecting the appropriate timescales over which to average the data is important to the strength of the relationship; 4) total nutrients were better correlated with biomass than dissolved inorganic nutrients; 5) watershed nutrient loads and ambient nutrient concentrations at the segment site were significantly correlated with one another on annual timescales; and 6)relationships between nutrient loads and aquatic primary producer biomass were only significant when estuarine volume and residence time are taken into account. While these positive relationships inspire confidence in the use macroalgae and phytoplankton biomass as indicators of eutrophication in SCB estuaries, these models are more reflective of the expression of eutrophication symptoms along a disturbance gradient. More in-depth studies in individual estuaries should be conducted before using the models to set site specific water quality goals to prevent or mitigate eutrophication.

In contrast to algae, extent of low dissolved oxygen events (measured as percent of time less than 5.7 mg L^{-1}) had no significant correlation with N and P loads; instead, it was significantly correlated with sediment organic carbon and total nitrogen content (Figure IV-7). Macroalgae was also significantly correlated with sediment organic carbon and total nitrogen (Figure III-11). Sediment organic matter content generally increases along a gradient of increasing eutrophication (Nixon 1995, Pelletier *et al*. 2010), with increased amounts of reflecting accumulation of allochthonous organic matter loading and autochthonous production within an estuary over decadal time-scales. Thus, dissolved oxygen and algae indicators integrate the effects of increased nutrient loading over very different time-scales. Macroalgae and phytoplankton biomass reflects a more immediate response to nutrient loads entering on that particular year, while low dissolved oxygen is largely driven by the combination of organic matter loading and algal biomass which has accumulated over time. These findings have important implications for how different response indicators can be used for management of eutrophication in estuaries. If nutrient loads are reduced, one may expect to see a response in algal blooms relatively quickly, while the recovery time required to address hypoxia problems in an estuary may be greatly extended.

In addition, strong feedback loops exist between sediments, surface waters, algae, and dissolved oxygen. Sediment organic matter content had a significant positive correlation with macroalgal biomass. Higher biomass of macroalgal mats sitting directly on the sediment can increase sediment organic matter content as the mat dies off and decays; sediments with high organic matter (and therefore nutrient) content can contribute to internal nutrient loading by recycling nutrients into the surface water; in some estuaries this sediment recycling can represent >80% of the total load of nutrients to surface waters. In shallow estuaries, sediments with high organic matter are a source of oxygen demand. However, high organic matter content is a natural feature of estuarine sediments, particularly in intertidal flats and marsh habitat, so thresholds for what may be considered eutrophic are not well accepted.

Recommended Next Steps

Create An Assessment Framework Appropriate For California Estuaries. The state of California is in the process of developing nutrient criteria to address eutrophication in estuaries. Results from the Bight'08 regional assessment can be utilized to inform this process by highlighting which indicators are relevant and how sensitive the results are to threshold selection, spatial and temporal sampling, as well as data management. Furthermore, the experience of the Bight estuary committee can be used to refine protocols to optimize monitoring for eutrophication by identifying trade-offs between more data and a better assessment. However, there are still a number of issues that must be addressed to create a scientifically defensible assessment framework for the state of California. These issues include protocol refinement, science supporting the selection of thresholds, and how to incorporate duration (length and frequency) of blooms and hypoxia. Furthermore, seagrass was not assessed in this survey and should be incorporated into future efforts to inform appropriate end-points for SCB estuaries with seagrass habitat.

Refine Predictive Load - Response Models. Analysis of the relationships between nutrient loading and ecological response in the Bight'08 study was limited to simple statistical models. While a relationship between algae and nutrient loads was identified, these models lacked precision and are not yet appropriate for management use to set site specific water quality goals. The predictive capability of these models can be improved through: 1) development of, at minimum, improved data and models of estuarine hydrology, shown to be critical in improving load-response relationships, 2) mechanistic studies of processes (e.g., denitrification, etc.) known to mitigate the effects of eutrophication. This information can be incorporated into a regional tool for scenario analysis of various nutrient loading rates and expected estuarine response.

APPENDIX A - B08 PARTICIPANTS

ftp://ftp.sccwrp.org/pub/download/DOCUMENTS/TechnicalReports/711_B08EE_AppendixA.pdf

APPENDIX B - QA/QC

ftp://ftp.sccwrp.org/pub/download/DOCUMENTS/TechnicalReports/711_B08EE_AppendixB.pdf

APPENDIX C - TECHNICAL SUPPORT FOR NUTRIENT CRITERIA DEVELOPMENT: SOUTHERN CALIFORNIA ESTUARINE EUTROPHICATION ASSESSMENT QUALITY ASSURANCE PROJECT PLAN

ftp://ftp.sccwrp.org/pub/download/DOCUMENTS/TechnicalReports/711_B08EE_AppendixC.pdf

APPENDIX D - INDIVIDUAL ESTUARY SUMMARIES

ftp://ftp.sccwrp.org/pub/download/DOCUMENTS/TechnicalReports/711_B08EE_AppendixD/

Agua Hedionda Lagoon (AHL)

Bolsa Chica Fully Tidal Lagoon (BCF)

Bolsa Chica Muted Tidal Lagoon (BCM)

Ballona Lagoon (BL)

Batiqutios Lagoon (BQL)

Ballona Wetlands (BW)

Devereux Lagoon (DL)

Goleta Slough (GS)

Los Peñasquitos Lagoon (LPL)

Mission Bay (MB)

Mugu Lagoon Fully Tidal (MLF)

Mugu Lagoon Muted Tidal (MLM)

Santa Ana River Wetlands (SAR)

Anaheim Bay/Seal Beach Fully Tidal (SBF)

Anaheim Bay/Seal Beach Muted Tidal (SBM)

Santa Clara River Estuary (SCR)

San Diego Bay Fully Tidal (SDF)

San Diego Bay Muted Tidal (SDM)

San Diego River (SDR)

San Elijo Lagoon (SEL)

San Juan Creek (SJC)

San Mateo Lagoon (SMC)

Santa Margarita Estuary (SME)

Topanga Canyon (TC)

Tijuana River Estuary (TJE)

University of California, Santa Barbara Campus Lagoon (UCL)

Zuma Canyon Lagoon (ZC)