A regional survey of the extent and magnitude of eutrophication in Mediterranean estuaries of southern California, USA

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Abstract

The magnitude and extent of eutrophication was assessed at 27 segments in 23 estuaries in the Southern California Bight (SCB) between October 2008-2009. We applied thresholds from the existing assessment frameworks from both the European Union (EU) and the US National Eutrophication Assessment to measurements of three indicators [macroalgae biomass and cover, phytoplankton biomass, and dissolved oxygen (DO) concentration] to categorize eutrophic condition in each estuary. Based on these frameworks, a large fraction of segments had moderate or worse eutrophic condition: 78% based on macroalgae, 39% for phytoplankton, and 63% for DO. Macroalgal biomass exceeding 70 g dw m^{-2} and 25% cover was found at 52% of sites during any sampling event. Thirty-three percent of segments exceeded this biomass for eight weeks or longer, a duration found to negatively impact benthic infauna. Duration of hypoxic events (DO <4 mg L^{-1}) was typically short, with most events less than one day; although 53% of segments had at least one event longer than 24 hours. Assessment frameworks of eutrophic condition are likely to evolve over time as the body of literature on eutrophication grows including aspects such as the applicability of indicators in specific habitat types, indicator thresholds, and how event frequency and duration are incorporated. This paper informs this debate by discussing how eutrophic conditions in SCB estuaries are categorized using different indicators and thresholds. To this end, categorization of estuarine eutrophic condition was found to be very sensitive to the type of threshold, how data are integrated to represent duration or spatial extent, and how indicators are used as multiple lines of evidence.

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Introduction

Eutrophication of estuaries is a global environmental issue, with demonstrated links between anthropogenic 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). Management of eutrophication and development of appropriate nutrient water quality goals is often hampered by a lack of regional monitoring data characterizing the symptoms, extent, and magnitude of the problem. A 2007 study of US estuaries conducted through the National Oceanic and Atmospheric Administration's (NOAA) National Estuarine Eutrophication Assessment (NEAA) found the majority of estuaries assessed had overall conditions rated as moderate to highly eutrophic (Bricker *et al.* 2008), yet highlighted significant data gaps. In the Southern California Bight (SCB), one of the most populated regions in the US, only eight of the region's 76 enclosed bay, lagoonal and river mouth estuaries were on the list of NEAA study sites and there was adequate data in only two of the eight SCB estuaries to assess eutrophic status. Among SCB estuaries, smaller "bar-built" lagoons and river mouth estuaries, which represent 22% of the areal extent but 82% by number in the region, are particularly data poor (Fong and Zedler 2000). These Mediterranean-type "bar-built" estuaries often experience restriction or complete closure to surface water tidal exchange due to the formation of sand-bars at their inlets (Webb *et al.* 1991, Largier *et al.* 1996). Consequently, they have increased susceptibility to eutrophication due to restricted flushing (Painting *et al.* 2007, Zaldivar *et al.* 2008). Additional data on the status of eutrophication in SCB estuaries are needed to determine the extent and magnitude of eutrophication in the region.

Over the past decade, much work has been done to establish standardized methodologies to assess eutrophication (Bricker *et al.* 2003, Zaldivar *et al.* 2008, Andersen *et al.* 2011, Devlin *et al.* 2011) and conduct surveys to evaluate the magnitude and extent of eutrophication (Bricker *et al.* 1999, Borja *et al.* 2009a, Andersen *et al.* 2011, Devlin *et al.* 2011, Garmendia *et al.* 2012). These assessment methodologies are the foundation worldwide for routine monitoring and establishment of water quality and biological objectives that are used to protect pristine habitat, identify impaired waterbodies, and provide targets for restoration or mitigation of systems where adverse effects of eutrophication have already occurred. Studies comparing assessment results generated for the same estuary have indicated that results vary slightly depending on which framework is applied (Devlin *et al.* 2011, Garmendia *et al.* 2012). Many of the frameworks apply similar indicators, but differences in time scales of data analysis (seasonal versus annual), characteristics included in the indicator metrics (concentration, spatial coverage, frequency of occurrence), and how to combine indicators into multiple lines of evidence, had an effect on the overall outcome of the assessment (Devlin *et al.* 2011). Most these assessment frameworks combine indicators of pressure (nutrient loads, estuarine surface water nutrients) with response indicators, often resulting in a numeric integrative index developed by expert best professional judgment (Ferreira *et al.* 2011). Evaluating the applicability of various indicators and respective thresholds from these frameworks and quantifying effect of data integration decisions is an important exercise to consider whether or how to adapt these assessment frameworks to use outside of the region in which they were originally developed.

The Mediterranean-type estuaries of the SCB are an excellent test case in which to conduct this evaluation. The Southern California Bight 2008 Regional Monitoring Program (Bight'08) 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. The objectives of the Bight '08 Eutrophication Assessment of the were to: 1) evaluate which indicators are relevant in Mediterranean estuaries such as those in the SCB; 2) explore how spatial and temporal integration of monitoring data affects the assessment of eutrophication status; and 3) estimate the extent and magnitude of eutrophication in SCB estuaries using appropriate indicators and thresholds. Results from this study can be used to inform the ongoing refinement of assessment approach and data integration decisions that affect the results of assessment of eutrophication in Mediterranean estuaries.

Methods

Study Area

The SCB (Figure 1) is an open embayment on the US West coast between Point Conception, California and Cabo Colnett (south of Ensenada, Mexico) and contains 76 estuaries ranging in size from 1 ha to over 50,000 ha. The SCB landscape overall is a

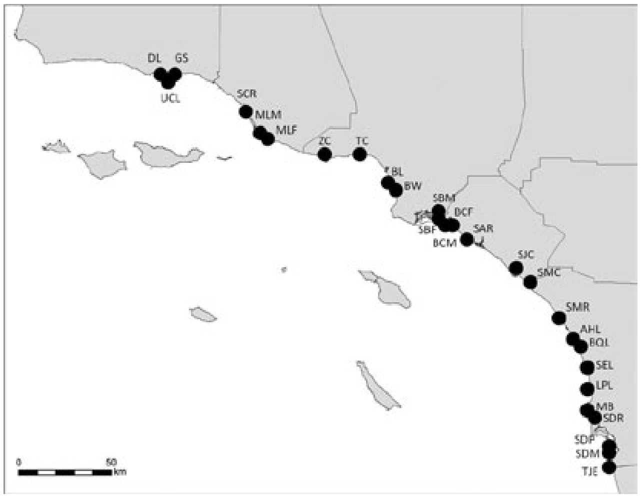

Figure 1. Map of Bight '08 Eutrophication Assessment Segment Sites.

highly developed urban environment; however, the watersheds within the SCB are highly variable in terms development. Watersheds ranged from a percent impervious surface of 1% (San Mateo Lagoon and Topanga Canyon Lagoon) to greater than 60% impervious (Anaheim and San Diego Bays). This 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). Agriculture was also prevalent in some watersheds ranging from 2% agricultural land use (Anaheim Bay) to 30% (Santa Margarita River Estuary). The SCB has a Mediterranean climate, with an average annual rainfall of 10 to 100 cm (Nezlin and Stein 2005), falling primarily during winter months (December through March), in approximately 20 annual storm events (Ackerman and Weisberg 2003). Winter runoff to the SCB contributes 95% of the total annual runoff volume and 67% of the total annual nitrogen and phosphorus loads (Ackerman and Schiff 2003, Sengupta *et al.* unpublished data).

Study Design

The Bight'08 Eutrophication Assessment was a synoptic study of eutrophication indicators, estuarine water column nutrient concentrations, and riverine nutrient loads monitored for one water year between October 2008 and October 2009. Estuaries were randomly selected from a comprehensive list of estuaries, proportional to the number of estuaries in each geoform: enclosed bay, lagoon or river mouth and tidal inlet status (open to tidal exchange, anthropogenically muted through tide gates, perennially or seasonally closed with no tidal exchange) in the northern portion of the SCB; all estuaries in San Diego County were included. Because primary producer expression is highly spatially variable within an estuary, we endeavored to make the results comparable across estuaries by selecting an index area, or "segment," which varies in size depending on the estuary (Table 1). The segments were proximal to the source of freshwater nutrient loading rather than the ocean inlet, representing a location within the estuary that is more likely exhibit symptoms of eutrophication. Segment sites were revisited each time period to determine seasonal variability in eutrophication indicators. Consequently, this survey represents a conservative assessment of eutrophication for each estuary and the region overall. A total of 27 segments were selected in 23 estuaries (Tables 1 and 2; Figure 1).

Estuaries are highly variable in how they respond to nutrient loading due to differences in site-specific controls on hydrology and other factors (Dettmann 2001, Pinckney *et al.* 2001, Zaldivar *et al.* 2008, Duarte 2009, Duarte *et al.* 2009). 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). Consequently, there has been a shift towards the use of eutrophic response indicators to estimate extent and magnitude of eutrophication (Bricker *et al.* 2003, Devlin *et al.* 2007, Zaldivar *et al.* 2008). Therefore, reporting of eutrophic condition in SCB estuaries was focused on the ecological response to nutrient over-enrichment rather than estuarine nutrient concentrations or watershed nutrient loads. For this assessment, data on a number of indicators were collected, though we opted to focus on dissolved oxygen and primary producer abundance because they have the most supporting data in the literature. Nutrient loads for each watershed are also reported for context.

Field and Laboratory Methods

Field methods consisted of three types of sampling: 1) continuous monitoring of dissolved oxygen, chlorophyll *a* fluorescence, temperature, salinity, pH and turbidity using a moored data sonde, 2) sampling of estuarine surface water and sediment nutrients, and primary producer biomass every other month, and 3) monitoring of storm and dry weather freshwater nutrient loads. Riverine nutrient loads were estimated

Table 1. Bight'08 Eutrophication Assessment estuary segments.

Name (Code)*	Latitude	Longitude	Watershed Area (km^2)	Estuary Area (m^2)	% of Estuary in Segment	Habitat Type				Max Salinity (ppt)	Min Salinity (ppt)	Annual TP Load (kg)**	Annual TN Load (kg)**
						Subtidal Unveg	Subtidal Eelgrass	Mudflat	Marsh				
Goleta Slough (GS)	34.4203	-119.8437	119	804,812	50%	20%	0%	12%	68%	57.6	35.4	8,518	20,369
Devereux Lagoon (DL)	34.4137	-119.8777	10	231,885	100%	41%	0%	27%	32%	59.6	18.1	3,198	7,287
UCSB Campus Lagoon (UCL)	34.4103	-119.8485	10	124,229	75%	96%	0%	0%	4%	55.9	46.9	222	1,257
Santa Clara River (SCR)	34.2317	-119.2626	4210	1,412,587	75%	50%	0%	48%	2%	3.2	1.6	22,483	239,378
Mugu Lagoon-Muted (MLM)	34.1098	-119.1407	803	14,098,340	25%	8%	0%	13%	79%	61.7	40.1	65,281	209,210
Mugu Lagoon-Full (MLF)	34.1033	-119.0927	803	14,098,340	15%	8%	0%	13%	79%	42.8	32.7	65,281	209,210
Topanga Canyon Lagoon (TC)	34.0386	-118.5831	51	3,678	100%	100%	0%	0%	0%	41.3	12.4	25	187
Zuma Canyon Lagoon (ZC)	34.0143	-118.8210	23	17,544	100%	56%	0%	13%	31%	7.3	0.6	154	578
Ballona Lagoon -Muted (BL)	33.9709	-118.4583	354	22,950	100%	71%	0%	29%	0%	54.4	45.6	13,939	95,098
Ballona Wetlands-Muted (BW)	33.9648	-118.4482	354	627,857	100%	0%	0%	19%	81%	48.7	46	13,939	95,098
Anaheim Bay/SealBeach-Muted (SBM)	33.7463	-118.0703	130	4,194,998	10%	28%	11%	5%	57%	55.3	51.3	550	13,832
Anaheim Bay/SealBeach-Full (SBF)	33.7354	-118.0756	130	4,194,998	50%	28%	11%	5%	57%	53.4	50.7	550	13,832
Bolsa Chica-Full (BCF)	33.7002	-118.0427	25	1,708,457	50%	62%	8%	21%	10%	53.4	51.4	25	2,500
Bolsa Chica-Muted (BCM)	33.6960	-118.0457	59	651,724	50%	72%	0%	7%	20%	55.2	49.8	1,081	5,840
Santa Ana R. Wetlands-Muted (SAR)	33.6378	-117.9547	4336	331,670	75%	37%	0%	14%	50%	52.7	51.1	16,528	89,981
San Juan Creek (SJC)	33.4628	-117.6832	458	64,839	100%	35%	0%	65%	0%	29.7	9.3	7,695	30,062
San Mateo Lagoon (SMC)	33.3865	-117.5937	346	126,262	100%	20%	0%	0%	80%	1.4	0.7	656	8,895
Santa Margarita Estuary (SME)	33.2346	-117.4090	1918	1,020,806	75%	33%	0%	24%	43%	51.7	13.7	19,177	137,799
Agua Hedionda Lagoon (AHL)	33.1421	-117.3284	546	1,410,480	50%	60%	17%	11%	12%	52.4	49.8	3,283	57,185
Batiquitos Lagoon (BQL)	33.0910	-117.3008	131	2,022,063	50%	18%	29%	20%	33%	51.6	49.3	46,966	60,037
San Elijo Lagoon (SEL)	33.0127	-117.2722	210	1,261,824	75%	9%	0%	54%	38%	47.3	17.2	2,080	63,262
Los Penasquitos Lagoon (LPL)	32.9350	-117.2604	244	1,306,657	50%	7%	0%	7%	85%	51.3	48.1	5,669	28,325
Mission Bay-Full (MB)	32.7720	-117.2138	118	8,795,281	15%	18%	75%	5%	2%	55.9	52.5	7,902	41,283
San Diego River (SDR)	32.7597	-117.2153	1120	1,142,328	50%	45%	0%	23%	32%	49.4	29.9	4,629	54,066
San Diego Bay-Muted (SDM)	32.6211	-117.1175	362	17,898,631	5%	28%	0%	71%	1%	71.1	63.2	8,045	147,537
San Diego Bay-Full (SDF)	32.5992	-117.1190	1037	50,094,111	10%	63%	22%	13%	2%	55.7	48.1	8,045	147,537
Tijuana River Estuary (TJE)	32.5683	-117.1313	4452	2,755,819	25%	7%	0%	17%	76%	52.7	46.2	82,988	348,018

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

**loads calculated from continuous flow data and discrete nutrient concentration measurements collected from the freshwater source.

Table 2. Summary of data integration options that were evaluated through sensitivity analyses. * designates options applied for the B'08 Eutrophication Assessment.

Issue	Indicators		
	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 Data Set	Transect values generated from: ***Quadrat averages** Quadrat percentile Quadrat maximum	Instantaneous ***Daily running average**	Instantaneous ***Hourly running average**
Spatial Extent for Data Integration	Segment values generated from: ***Average of Transects** Maximum Transect Percentile of Transect Data	***Single Location**	***Single Location**

using a combination of field measurements and modeling.

Riverine Nutrient Loads

Total riverine nutrient fluxes to the SCB were estimated using empirical wet and dry weather data for monitored watersheds in combination with modeled wet weather fluxes for unmonitored watersheds. Continuous discharge was measured with flow gauges at some sites and from ratings curves based on continuous water level (recorded with a HOBO data logger, Onset Corp.) and quarterly measurements of flow and channel topography. Discrete total nitrogen (TN) and phosphorus (TP) samples were collected for wet and dry weather (October 2008-2009), and analyzed via persulfate digest (Patton and Kryskalla 2003) and analyzed as colorimetrically using an autoanalyzer. The spreadsheet model originally developed by Ackerman and Schiff (2003), based on the Rational Method, was updated and modified to predict TN and TP loads using updated land use-specific runoff concentrations for nutrients (Howard *et al.* 2012, Sengupta *et al.* unpublished data).

Dissolved Oxygen and Water Column Physio-Chemistry

Water column physiochemical parameters and water level were measured continuously using a YSI 6600 data sonde from January through October 2009. 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 (30 cm from the sediment surface). Measurements were collected every 15 minutes and an hourly running average was applied to the data set.

Macroalgae Biomass and Cover

Macroalgal biomass and cover was measured at three 30 to 50 m transects in the lower intertidal zone (Kennison *et al.* 2003). Percent cover was measured at ten randomly allocated points along each transect using the point intercept method with 0.5 m^2 quadrats. Biomass was comprehensively collected at five of the quadrat locations from a prescribed surface area. Biomass samples were stored at 4°C and processed within 24 hours of collection. In the laboratory, algal samples were cleaned of macroscopic debris, mud and animals. Excess water was shed from each sample, weighed wet, dried at 60°C to a constant weight, then weighed dry.

Phytoplankton Biomass

Phytoplankton biomass was estimated from fluorescence measurements collected via in situ optical probe (YSI 6600 sonde, chlorophyll fluorescence probe), and discrete chlorophyll *a* water grab samples taken every other month. Water column chlorophyll *a* samples were filtered on a Whatman GF/F and frozen for subsequent analysis using EPA 445 protocols on a Turner Designs fluorometer within 28 days of collection. In situ chlorophyll fluorescence probes were maintained according to factory specifications and were routinely calibrated. Fluorescence measurements were calibrated to chlorophyll *a* concentrations using least squares regression of daily averaged data probe measurements and discrete concentration data collected on the same day. The least squares fit had R^2 values ranging from 0.413 to 0.995 with an average of 0.747 and 89% of sites had an R^2 greater than 0.5. The poor fit for some sites is likely related to the disparity between where the measurements were collected (surface waters for discrete samples, bottom water for chlorophyll fluorescence probe). Most sites were shallow and the depth difference was insignificant, but for the few deeper sites, it seemed to impact fit.

Eutrophication Indicators

Several assessment frameworks have been developed to assess eutrophic condition of estuaries utilizing a range of indicators (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, to capture frequency of occurrence and spatial extent in indicator metrics, and use a combination of indicators into an overall condition rating (Devlin *et al.* 2011). For this study, we selected individual indicators from established assessment frameworks to evaluate how well they worked in SCB estuaries. Indicators were evaluated from the European Union - Water Framework Directive (EU-WFD) and the US Assessment of Estuarine Trophic Status (ASSETS). The EU-WFD was developed to regulate and monitor water bodies in EU member countries, organizing management of waterbodies by catchment and standardizing protocols across Europe (Borja *et al.* 2006, Hering *et al.* 2010). Several assessment frameworks are associated with the EU-WFD (Ferreira *et al.* 2011, Birk *et al.* 2012); we selected two of these that utilized indicators prevalent in SCB estuaries: French Research Institute for Exploration of the Sea (IFREMER) and the United Kingdom WFD protocols (UK-WFD). ASSETS (Bricker *et al.* 2003) was developed to assess the status of eutrophication in US estuaries through NOAA's National Estuarine Eutrophication Assessment (Bricker *et al.* 1999). These frameworks assess response indicators, rather than ambient physical or chemical variables alone, although pressure variables such as nutrient loads and concentrations are included in the overall assessment (Borja and Dauer 2008, Borja *et al.* 2011b).

Because the estuaries in this study were not comprehensively characterized for pressure and susceptibility factors, which are required for an overall assessment of eutrophic condition in these frameworks, we conducted an analysis of eutrophication based on measurements of response indicators prevalent in SCB estuaries to develop a regional estimate of extent and magnitude of eutrophication in the SCB. Furthermore, we examined the sensitivity of the results to changes in threshold selection, data format, and data integration. Eutrophic condition category was assigned to each segment for each indicator (generating a set of assignments for each estuary segment). Details of how monitoring data were used to calculate final segment categorization is given by indicator below.

Macroalgal Abundance

A modification of the UK WFD element for macroalgae based on a combination of biomass and cover was applied (Scanlan *et al.* 2007). Thresholds for wet weight biomass were converted to dry weight utilizing the median dry:wet weight ratio of all ulvoid biomass samples. These thresholds were used to determine condition using the average biomass and cover scores from the two consecutive periods of highest biomass and cover (Peak Season; Figure 2a), though other data integration options were explored (Table 2).

Phytoplankton Biomass

Phytoplankton assessment as an indicator were available from ASSETS (Bricker *et al.* 2003) and from the French Research Institute for Exploration of the Sea in the EU (IFREMER; Souchu *et al.* 2000, Zaldivar *et al.* 2008; Figure 2b). We chose to apply the IFREMER framework because French Mediterranean lagoons were expected to be similar to SCB estuaries. Thresholds were applied to the annual average of chlorophyll *a* data, though other data integration options were explored (Table 2).

a) **UK-WFD numeric thresholds for assessing eutrophic condition using macroalgae biomass and cover (adapted from Scanlan *et al.* 2007).**

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

b) **IFREMER (Souchu *et al.* 2000) and ASSETS (Bricker *et al.* 2003) numeric thresholds for assessing eutrophic condition using phytoplankton biomass (as suspended chlorophyll *a*).**

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

c) **UK-WFD (Best *et al.* 2007) and ASSETS (Bricker *et al.* 2003) numeric thresholds for assessing eutrophic condition using dissolved oxygen concentration.**

Framework	Proposed Eutrophic Status	Freshwater	Marine	Rational
EU-WFD	High	≥7 mg L^{-1}	≥5.7 mg L^{-1}	Supports all□life stages of salmonids and transitional fish
	Good	5 - 7 mg L^{-1}	4.0 - 5.7 mg L^{-1}	Supports □resence 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
	Poor	2 - 3 mg L^{-1}	1.6 - 2.4 mg L^{-1}	Supports non-salmonid presence, poor survival salmonids
	Bad	<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 response in invertebrates and fish
	Hypoxia	0 - 2 mg L^{-1}		Significantly reduced benthic macroinfauna, epifauna and demersal species

*EU-WFD scores are "High" = nearly undisturbed conditions; "Good" = slight change in composition and/or biomass; "Moderate"= moderate change in composition and/or biomass; "Poor" = major change in biological communities; "Bad" = severe change in biological communities.

** Moderate or worse status is considered actionable for the EU-WFD; estuaries must have a good or high condition.

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

Figure 2. Assessment frameworks used for evaluating eutrophic condition in SCB segments.

Dissolved Oxygen Concentration

Dissolved oxygen assessment frameworks from ASSETS and the UK-WFD are based on adverse effects of hypoxia on benthic and demersal fauna (Figure 2c). However, the proposed framework for the UK-WFD (Best *et al.* 2007) incorporates the effect of salinity on oxygen solubility by shifting the threshold based on the measured salinity. We opted to use a hybrid of the two approaches by applying the UK-WFD thresholds to the 10th percentile of annual data as prescribed by ASSETS (90% of data exceeds DO thresholds; Figure 2c), though other data integration options were explored (Table 2). The UK-WFD thresholds are comparable to those generated independently for susceptible California fish and invertebrate species (Sutula 2011), but use of the fifth percentile was designed for use in well ventilated waters and we measured DO in bottom waters that were often stratified. Because the ASSETS thresholds are applied to the 10th percentile of bottom water DO, a hybrid approach was considered appropriate in this case.

Results

Extent and Magnitude of Eutrophication in SCB Estuaries

Macroalgal Abundance

Seventy-eight percent of segments had moderate or worse eutrophic condition with respect to macroalgae based on application of the UK-WFD macroalgal indicator applied to peak season biomass and cover (Figure 3a). Estuaries in the "moderate" category (37%) had cover between 25% and 50% and biomass less than 70 g dw m^{-2} (Figure 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, DL) and 91 g dw m^{-2} and 93% cover (site with highest cover, GS). The number of consecutive periods any segment spent in each eutrophic condition category was variable from site to site (Figure 3c). Some segments had chronically high biomass and cover and were classified as moderate or worse for more than 80% of the year (e.g., DL, GS, UCL, MLM, BL), whereas other sites had episodic blooms measured during a single period (e.g., ZC, SJC, SBF). For most segments, the overall score was driven by more than one period of moderate or worse eutrophic condition.

Fifteen segment sites (55%) had macroalgae biomass and cover indicative of moderate or worse eutrophic condition for two or more consecutive periods (>8 weeks). Thirty percent of segments had two or more periods of poor/bad eutrophic condition and 11% of the segments had two or more consecutive periods of bad eutrophic condition. Thirty-seven percent of segments 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%) had continuous coverage of moderate or worse eutrophic condition throughout the sample year and one of these sites was continuously in a poor or bad condition (UCL).

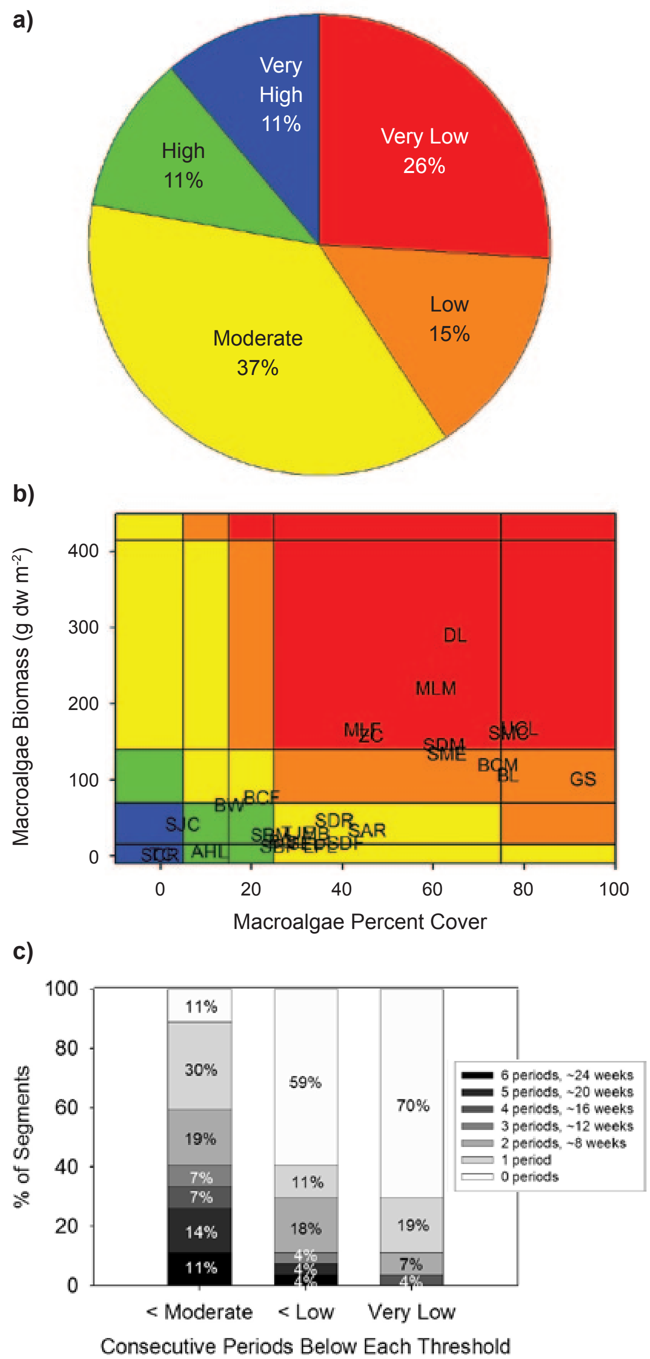

Figure 3. Percent of segments falling into each eutrophic condition category based on macroalgae biomass and cover (a), macroalgae biomass and cover values for each SCB segment (b), and duration of macroalgae bloom for each segment (c).

Phytoplankton Biomass

Annual average chlorophyll *a* concentrations ranged from 0.5 to 42 µg L^{-1}. Of the three biological response indicators, phytoplankton biomass had the fewest number of segments categorized in moderate or worse eutrophic condition (39%; Figure 4a). Eleven percent of segments scored in the "moderate" category. SCR and SJC had little macroalgae (categorized as “very high eutrophic condition” based on macroalgal abundance), but high chlorophyll *a* (categorized as “bad” for SCR and “poor” for SJC).

Daily eutrophic condition was also categorized for each segment to determine duration of phytoplankton bloom events. One third of segments spent less than 10% of the time in an eutrophic condition category of moderate or worse (90% of the time in eutrophic condition of good or very high according to the IFREMER thresholds). Some segments had chronically high chlorophyll *a* classified defined by having moderate or worse condition for over half of the year (e.g., SCR, SDM, SJC, SDR, MLM), whereas other sites had more episodic blooms lasting only a few weeks (e.g., BW, SBM, SBF, BCM, BCF, SEL, MB).

All segments categorized with moderate or worse overall eutrophic condition had daily average chlorophyll *a* concentrations greater than 7 µg L^{-1} for more than 25% of the year. With respect to bloom duration, 40% of segments had continuous phytoplankton blooms greater than 7 and 10 µg L^{-1} for longer than one month (Figure 4b). Twenty-five percent of segments had chlorophyll *a* greater than 7 µg L^{-1}, and 22% greater than 10 µg L^{-1}, for longer than two months. Fifteen percent of segments had chlorophyll *a* greater than 30 µg L^{-1} for one month and 8% for two months. Some systems with annual average chlorophyll *a* indicative of “good” or “high” eutrophic condition, had concentrations above 7 µg L^{-1} for periods less than one month in duration.

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 do not likely impact the overall condition category. 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. Phytoplankton blooms in SCB estuaries were typically in late spring/early summer; thus, the data gap for these two sites may have resulted in a lower condition category than if the sondes had been deployed for the full period.

Dissolved Oxygen

For dissolved oxygen (DO), 10^{th} percentile concentrations over the nine-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

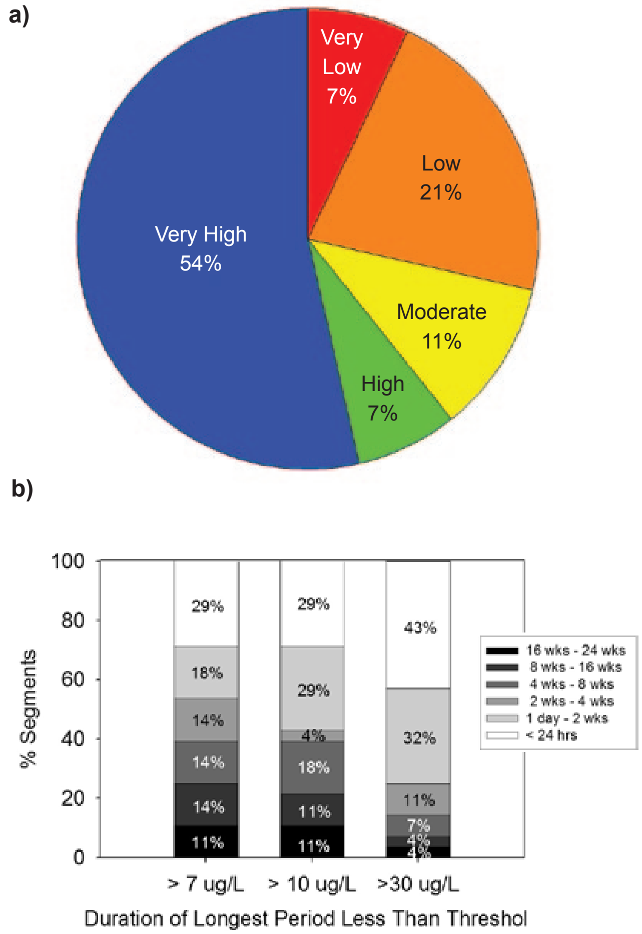

Figure 4. Percent of segments falling into each eutrophic condition category based on chlorophyll *a* (a) and duration of phytoplankton bloom events in SCB segments (b).

eutrophic condition category of moderate or worse using the UK-WFD thresholds (Figure 5a), and of these systems, half were categorized with “bad” eutrophic condition (36% of all segments). Thirty-nine percent of segments had good or high eutrophic condition protective of adult salmonid survival, and 11% of segments fell in the high condition category, which protects all life stages of salmonids.

Eutrophic condition at each segment was also categorized continuously for each segment using the hourly running average of 15-minute DO concentrations to determine the duration of hypoxic events. The percentage of time any segment spent in each eutrophic condition category was variable (Figure 3c). Most segments fall into the moderate or worse condition for a portion of the diel cycle (night), and the overall percentage of time in a moderate or worse eutrophic condition (as well as the tenth percentile value) is reflective of many consecutive nights of low

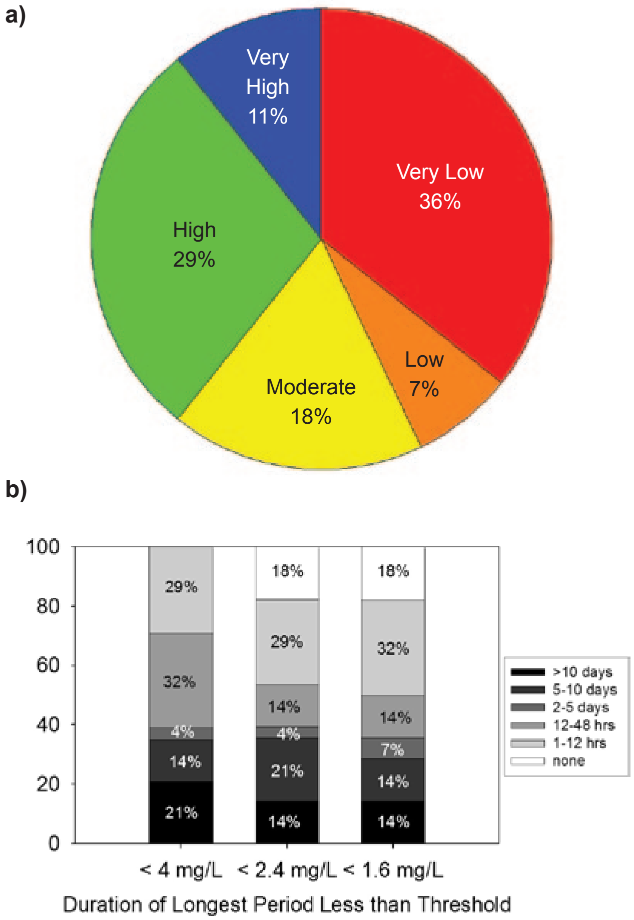

Figure 5. Percent of segments in each condition category based on DO (a) and duration of hypoxic events in SCB segments (b).

dissolved oxygen concentration rather than a single continuous period of low DO. However, for some segments, continuous low DO events exceeded diel cycles (e.g., SCR, DL, GS, UCL). All segments had some period less than the moderate threshold of 4 mg L^{-1}, and 82% of sites spend some time below the poor and bad thresholds (2.4 mg L^{-1} and 1.6 mg L^{-1} respectively). The longest continuous period less than the moderate and poor thresholds was 12 hours or less for 29% (Figure 5b). For longer duration events, 35% of segments had DO less than 4 and 2.4 mg L^{-1} longer than five days. Twenty-eight percent of SCB segments had DO continuously less than 1.6 mg L^{-1} for longer than 5 days and 14% for longer than 10 days (GS, SCR, SDR, UCL).

As with phytoplankton, data gaps in the continuous DO dataset were a concern for some segments, particularly MB and SDR due to the delay in deployment described above. For most segments, hypoxia occurs in late spring/early summer; thus, the data gap for these two sites could result in a lower eutrophic condition category than would have been achieved had the sondes been deployed for the full time.

Multiple Lines of Evidence

The three indicators of biological response to eutrophication did not necessarily agree in many SCB segments (Table 3). All but one segment (96% of segments) were assigned an eutrophic condition class of moderate or worse based on at least one indicator. 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 [a primary and secondary indicator as prescribed by the ASSETS framework (Bricker *et al.* 2003)]. Fifteen percent of segments (MLM, SDR, SMR, UCL) fell in a category of moderate or worse for all three indicators.

Segments in a Regional Context

We ranked segments from highest eutrophic condition to lowest (Table 3). The five highest ranked estuary segments in the SCB were BQL, SBF, LPL, BCF, and MLF. The five lowest ranked segments were MLM, SDR, UCL, SCR, and DL. However, in a number of segments, indicators often gave conflicting results. For example SCR and SJC ranked among the segments with highest eutrophic condition for macroalgae, but among the lowest for phytoplankton and low overall. Similarly, some sites ranked among the lowest condition based on macroalgae and highest for phytoplankton (BCM, BL). Most sites that ranked as low eutrophic condition for DO were also ranked as low eutrophic condition for one or both primary producers. However, there are also two sites that rank among the lowest eutrophic condition for both primary producers but among the highest for DO (MLF, SMC), though

Sensitivity of Results to Threshold, Data Format and Spatial and Temporal Integration

A number of segments were on the borderline of thresholds that would place them in a different category, making them prone to reclassification given a different data management regime or slight change in threshold. This would be particularly important for segments in the "moderate" category, since the threshold between "good" and "moderate" drives management action according to the EU-WFD. To investigate the sensitivity of eutrophic condition category to changes in data format, data

Table 3. Ranks of segments. Annual total nitrogen load is ranked from 1 (lowest load) to 27 (highest load).

	Overall Rank	Segment	Annual TN Load Rank	Segment Rank by Indicator*		
				Peak Season Macroalgal Abundance	Annual Mean Phytoplankton Biomass	10th Percentile Dissolved Oxygen
Highest Condition	1	Batiquitos Lagoon (BQL)	16	7	10	3
	2	Seal Beach (SBF)	9	5	12	4
	3	Los Penasquitos Lagoon (LPL)	11	6	7	10
	4	Bolsa Chica (BCF)	4	15	11	5
	5	Mugu Lagoon (MLF)	24	18	19	1
	6	San Diego Bay (SDF)	22	10	15	7
	7	Santa Ana R. Wetlands-Diked (SAR)	18	14	2	9
	8	Seal Beach- Diked (SBM)	8	9	8	14
	9	San Elijo Lagoon (SEL)	17	8	5	17
	10	San Mateo Lagoon (SMC)	7	24	18	2
	11	Topanga Lagoon (TC)	1	2	13	16
	12	Tijuana River Estuary (TJE)	27	11	1	19
	13	San Diego Bay- Diked (SDM)	23	22	24	6
	14	Agua Hedionda Lagoon (AHL)	15	3	20	11
	15	Ballona Lagoon- Diked (BL)	19	19	3	13
	16	Santa Margarita Estuary (SME)	21	20	21	12
	17	San Juan Creek (SJC)	12	4	25	8
	18	Zuma Canyon Lagoon (ZC)	2	17	14	18
	19	Bolsa Chica- Diked (BCM)	5	21	4	15
	20	Ballona Wetlands (BW)	20	13	6	26
	21	Mission Bay (MB)	13	12	9	27
	22	Goleta Slough (GS)	10	23	17	23
	23	Mugu Lagoon – Diked (MLM)	25	26	26	20
	24	San Diego River (SDR)	14	16	23	24
	25	UCSB Campus Lagoon (UCL)	3	25	22	22
	26	Santa Clara River (SCR)	26	1	27	21
Lowest Condition	27	Devereux Lagoon (DL)	6	27	16	25

integration, and threshold selection, we compared the results from the assessment as described above to results generated using a different data management options and thresholds from the ASSETS framework (Table 4).

For macroalgae, assessment outcome was sensitive to data format, and spatial and temporal integration of the data (Table 4). Use of wet weight versus dry weight had a significant effect on condition category, with 19% segments "improving" eutrophic condition category and 11% "declining". Categorizing a segment based on the transect with the highest biomass and cover, rather than the average of the three transects during the period of maximum biomass, decreased the eutrophic condition category for 44% of segments. Eleven percent of segments changed by two or more condition categories in this worst case scenario. Similarly, if an annual average of all segment data was used, 41% of segments increased in eutrophic condition category and one segment decreased. If the average of all transect data during the period of highest biomass and cover (maximum period) was used, 7% of segments increased in condition category and 22% decreased. Use of an annual average segment value generated from an average of the three transects resulted in the maximum number of segments being in the highest possible eutrophic condition category, although 56% of segments would not change. Use of the single period of highest biomass and cover (maximum period) generated from the transect with highest biomass and cover (maximum transect) resulted

Table 4. Changes in eutrophic condition class due to data format, framework, or data integration. PS = Peak Season. MP = Maximum period. Avg = Average.

Indicator and Comparator	Data Format	Framework	Data Management	# of 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	EU-WFD	Annual Avg/Transect Avg	15	9	2	1	0
		EU-WFD	PS Avg/ Transect Max	15	0	0	9	3
		EU-WFD	MP/ Transect Avg	22	0	0	4	1
		EU-WFD	MP/ Max Transect	10	0	0	13	4
	Wet Biomass	EU-WFD	Annual Avg/Transect Avg	14	8	4	1	0
		EU-WFD	PS Avg/ Transect Avg	19	5	0	3	0
		EU-WFD	PS Avg/ Transect Max	12	2	0	10	3
		EU-WFD	MP/ Transect Avg	19	2	0	6	0
		EU-WFD	MP/ Max Transect	10	0	0	12	5
Phytoplankton IFREMER Framework Annual Average of Daily Averages	Daily Averages	EU-WFD	75th %tile	22	1	0	5	0
		EU-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	EU-WFD	Annual Avg	27	0	0	1	0
		EU-WFD	75th %tile	23	3	0	2	0
		EU-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	EU-WFD	Annual Avg	16	2	4	3	2
		EU-WFD	75th %tile	17	2	3	0	5
		EU-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	EU-WFD	5th %tile	20	0	0	8	0
		EU-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	EU-WFD	5th %tile	20	0	0	8	0
		EU-WFD	10 %tile	28	0	0	0	0
		EU-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

in the lowest possible condition category for each segment, although 37% segments would not change.

Phytoplankton biomass results were sensitive to temporal integration of the data as well as thresholds (Table 4). The difference between using daily averages versus the instantaneous data set did not have a large effect on the outcome. However, there was an effect of using discrete data versus continuous data; 22% of segments increased in eutrophic condition category and 19% decreased when discrete data were used compared to continuous daily averages. Changing the data integration period also had a significant effect on the outcome. Using a 75th or 90th percentile instead of the annual average resulted in 22 and 44% of segments changing eutrophic condition class, respectively. Using ASSETS as described (thresholds applied to 90th percentile of annual data), 7% of segments increased in condition category and 30% decreased in category relative to IFREMER thresholds applied to annually averaged data.

Dissolved oxygen assessments were sensitive to changes in temporal integration and assessment framework (Table 4). Use of the 5th percentile resulted in eight segments (30%) scoring lower, whereas use of the 15th percentile resulted in 22% of segments scoring higher in eutrophic condition than the 10th percentile. 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 condition category. Changing the data format from an hourly running average to instantaneous generally had no effect, with the exception of the ASSETS framework, in which 37% of segments changed class when the hourly running average was used, with roughly equivalent numbers "improving" and "declining" condition class.

Discussion

Extent and Magnitude of Eutrophication in SCB Estuaries

Regional Condition

Eutrophication was found to be pervasive in SCB estuarine segments during the 2008-2009 water year (having an EU-WFD eutrophic condition category of moderate or worse) regardless of whether indicators were applied individually (78% based on macroalgae, 39% for phytoplankton, and 63% for DO), or as a part of a multi-metric approach (53% based on one primary producer and DO). The EU-WFD applies a "one out, all out" approach in determining eutrophic 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, all but one of 27 segments assessed would require management action to improve eutrophic condition. However, several studies have demonstrated that a multi-metric approach provides a more robust accounting of condition (Borja *et al.* 2009a,b, 2011b; Borja and Rodriguez 2010). The ASSETS framework is such an approach wherein the scores for primary symptoms (primary producer response) and secondary symptoms (DO) are combined to generate an overall score of eutrophic condition for the estuary (Bricker *et al.* 2003, Nobre *et al.* 2005). The applicable "primary symptom" would vary depending on the estuary and may vary from year to year (Cloern and Nicols 1985, Nixon *et al.* 2001, Sousa-Dias and Melo 2008). For example, two SCB estuaries had little macroalgae, but high suspended chlorophyll *a*, indicating that eutrophic condition in these systems is driven by phytoplankton. This highlights the importance of selecting the most critical primary producer response indicator for each system and each year, rather than a one size fits all approach (Bricker *et al.* 2003, Zaldivar *et al.* 2008, Borja *et al.* 2009b). Applying the worst primary symptom with dissolved oxygen as the secondary symptom to this assessment resulted in 53% of segments requiring management action to improve eutrophic condition to a "good" status.

Event Duration

Macroalgal and phytoplankton blooms in the SCB were of sufficient duration to impact benthic and pelagic fauna in some segments. Several studies have shown negative impacts from moderate levels of macroalgae biomass on benthic communities after eight to 20 weeks of exposure (Norkko and Bonsdorff 1996, Bolam *et al.* 2000, Cardoso *et al.* 2004, Cummins *et al.* 2004). Fifty-eight percent of SCB segments sampled had moderate or worse macroalgae biomass for eight or more weeks and 26% for longer than 20 weeks. For phytoplankton, blooms of short duration are vital to sustain estuarine food-webs (Cloern 1996, Cloern and Jassby 2008); however, blooms lasting longer than one to two months will begin to have a negative impact on submerged aquatic vegetation, decreasing habitat diversity and impacting eutrophic condition (Moore and Wetzel 2000, Ruiz and Romero 2001). Within the SCB, 19%

of segments had continuous phytoplankton greater than 10 µg L^{-1} for longer than two months. Fifteen percent of segments had biomass greater than 30 µg L^{-1} continuously for one month and 7% for two months. Thus, it is reasonable to assume a quarter or more SCB segments have sufficient macroalgae and/or phytoplankton bloom duration to significantly affect ecosystem health.

The length and frequency of hypoxia in SCB estuaries was also a concern. The response of aquatic organisms to low DO will depend on the intensity of hypoxia, duration of exposure, and the periodicity and frequency of exposure (Rabalais and Harper 1992). All SCB segments had a period less than the moderate threshold of 4 mg L^{-1}, and 82% of segments below 2.4 mg L^{-1}. However, for 29% of segments, the longest continuous period less than the moderate and poor thresholds was less than 12 hours, a length of time that can be endured by most organisms (Vaquer-Sunyer and Duarte 2008). Frequent hypoxic periods of short duration are typical of shallow subtidal or intertidally dominated habitats, where DO concentrations are driven by high sediment oxygen demand (Diaz *et al.* 1992, Rabalais *et al.* 1994, Sohma *et al.* 2008). While nightly hypoxia may not exceed the duration for lethal effects on many estuarine species, it can create chronic stress on animals, adversely affecting feeding, capacity to escape predation, reproduction and growth. Thirty-five percent of segments had DO concentrations less than 4 mg L^{-1} for longer than five days, the median lethal time upon exposure to hypoxia (Vaquer-Sunyer and Duarte 2008). This suggests that a third or more systems may have negative ecosystem effects due to low dissolved oxygen concentrations.

Comparison to Other Regions

The observed predominance of moderate to hyper-eutrophic condition in SCB estuaries is similar to other regional or national studies of US estuaries, as well as in Europe and Australia. Widespread coastal eutrophication has been reported for estuaries in the United States (NEEA), but prior to this study, the status of southern California estuaries was largely unknown (Bricker *et al.* 1999, Bricker *et al.* 2008). The majority of NEEA estuaries showed signs of eutrophication, with 65% displaying at least one symptom and 78% of assessed estuarine area falling into moderate or worse eutrophic condition conditions (Bricker *et al.* 1999, 2008). Several studies of eutrophication throughout the European and Australian coastlines have found that symptoms of eutrophication were present in half or more estuaries assessed (Hillman *et al.* 1990; Ærtebjerg *et al.* 2001; Borja *et al.* 2004, 2009a; Ferreira *et al.* 2007).

Uncertainty in the Assessment and Applicability of Existing Frameworks

The confidence with which managers will pursue remediation of a problem is dependent on the level of uncertainty in the assessment. Our data can be used to discuss the applicability of existing assessment frameworks and provide insights into improvements and adaptations to suit a wide range of estuaries. Uncertainty in the assessment can arise from several factors: 1) appropriateness of indicators; 2) how well the assessment captured the temporal and spatial variability; 3) applicability of assessment framework thresholds; and 4) how the data were used to categorize estuaries. These uncertainties are explored below and are illustrated in Table 4.

Adequacy of Indicators

Many studies support the use of macroalgae, phytoplankton and DO as indicators to assess eutrophication. Macroalgae was the dominant aquatic primary producer in most SCB estuaries and was particularly well suited to shallow intertidally dominated estuaries. Phytoplankton biomass and DO are most applicable in estuaries dominated by sub-tidal habitat and are less relevant in estuaries dominated by intertidal habitat. Twenty-six percent of SCB estuaries have more intertidal area than subtidal area. Therefore, although DO and phytoplankton thresholds were applied to all segments, they may not be relevant in all. However, clear guidance for when phytoplankton or DO should no longer be applied is generally not available and research to support development of guidance is limited.

Uncertainty from Temporal and Spatial Variability

Time and space matter when monitoring indicators for assessment of extent and magnitude of eutrophication and expression of eutrophication can be spatially and temporally variable (Cloern and Nicols 1985, Diaz *et al.* 1992, Rabalais *et al.* 1994, Nixon *et al.* 2001, Nobre *et al.* 2005, Sousa-Dias and Melo 2008, Nezlin *et al.* 2009, Vaquer-Sunyer and Duarte 2011). Furthermore, nutrient loading into estuaries can differ dramatically from year to year (Nixon *et al.* 2001, Kemp *et al.* 2005, Gilbert 2010), and this inter-annual variability will greatly affect expression of eutrophication symptoms (Pinckney *et al.* 2001).

This study adequately captured seasonal variability, though not interannual variability. The proposed EU-WFD framework recommends monitoring be conducted in at least three out of the five year reporting cycle (Best *et al.* 2007, Scanlan *et al.* 2007, Zaldivar *et al.* 2008). The 2008-2009 water year was relatively dry; nitrogen and phosphorus loading to SCB estuaries was in the 16th percentile of a 13-year estimate of nutrient loads (Howard *et al.* 2012). Thus, it is expected that the study data were representative of below average conditions.

Spatial heterogeneity is also characteristic of estuaries, so our use of a targeted index area introduces uncertainty in our ability to report on extent of eutrophication for each estuary individually. However, in roughly half of the estuaries, the segment represents 75% or more of the total estuarine area because SCB estuaries are typically small (<40 ha). Spatial variability within the segment was better accounted with macroalgae, which relied on data from three transects, distributed throughout the segment. However, phytoplankton and dissolved oxygen were only monitored at a single location.

Adequacy of Assessment Frameworks

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) have recognized that the lack of data 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, and how event duration and frequency are incorporated, are likely to change over time as the body of literature grows (Patricio *et al.* 2007, Scanlan *et al.* 2007, Domingues *et al.* 2008). One objective of this study was to inform this debate by discussing to what degree these frameworks were applicable to SCB estuaries and the associated uncertainties in their application.

For SCB estuaries, macroalgae is a key indicator for extent and magnitude of eutrophication. The Scanlan *et al.* (2007) macroalgal assessment framework accounts for both the abundance (biomass) and spatial patchiness (cover) inherent in this indicator. Results of a recent study of two California estuaries by Green *et al.* (In press) in Bodega Bay and Newport Bay show significant impacts on benthic invertebrates at 110 to 120 g dw m^{-2} and 100% cover after four weeks of constant biomass. Similarly, Bona (2006) showed an effect threshold on benthic habitat quality at biomass levels greater than 700 g ww m^{-2} (~ 90 g dw m^{-2}) and >70 % cover. Therefore, an "effects" threshold in the range of 70 to 120 g dw m^{-2} [as proposed by Scanlan *et al.* (2007)] seems reasonable. In a recent study, a "natural background" abundance of macroalgal biomass was quantified in the range of 2 to 16 g dw m^{-2} (Sutula *et al.* unpublished data), similar to the range of very high (0 - 10 g dw m^{-2}) established by best professional judgment in the EU WFD (Scanlan *et al.* 2007). At what areal percent cover this threshold is applied is another question. Diversity and biomass of epifauna was shown to increase with biomass until macroalgae covered 50% of the benthos (Pihl *et al.* 1996, 1999). Jones and Pinn (2006) found that after a month of approximately 75% macroalgal cover, all species in the sediment declined and many organisms started migrating out of the sediment and moving into the mats. However biomass was not monitored in these studies. In the SCB, placement of 10 segments (37%) in the "moderate" eutrophic condition was driven by cover between 25 and 50% with biomass less than 70 g dw m^{-2}. Placement of these segments in an "actionable" category may be overly conservative.

ASSETS (Bricker *et al.* 2003) and IFREMER (Souchu *et al.* 2000) thresholds for phytoplankton biomass are based on the paradigm of light limitation of benthic primary producers, particularly seagrass, although references to other adverse effects are made. However, this paradigm is not necessarily relevant in all systems. In the SCB, 26% of estuaries had seagrass habitat and 36% had brackish water submerged aquatic vegetation. Both ASSETS and IFREMER assessment frameworks have similar "no effect" levels of chlorophyll *a*: less than 5 µg L^{-1} and 7 µg L^{-1}, respectively. Moderate effects range from roughly 7 to 10 µg L^{-1}; similar to the criteria established for Yaquina Bay in Oregon, 3 - 5 µg L^{-1} (Brown *et al.* 2007), and Florida estuaries, <3.8 - 11.0 µg L^{-1} (Janicki *et al.* 2000, 2009). Above 20 µg L^{-1}, submerged aquatic vegetation show declines (Stevenson *et al.* 1993) and phytoplankton community shifts from diverse mixture to monoculture (Twilley 1985). 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). Estuaries with closed inlets are typically brackish and can become dominated by cyanobacteria under high nutrient loading (Paerl 2008); for these estuaries, studies of the relationships between chlorophyll *a* and cyanobacteria blooms can be illustrative (Walker 1985, TetraTech 2006). Cyanobacteria blooms are rare when summer mean *chlorophyll a* concentrations are less than 5 to 10 $\mu g\ L^{-1}$. These values are comparable to "no effect" levels in seagrass dominated habitats as described by ASSETS and the IFREMER. Similarly, concentrations of 20 $\mu g\ L^{-1}$ suggests cyanobacteria blooms will occur about 15 to 20% of the time (Walker 1985). Thus, while there are a few studies that provide a clear picture of biomass dose versus eutrophic response for phytoplankton, there appears to be some scientific consensus around ranges of thresholds (Borja *et al.* 2011a).

This study found that the use of the WDF framework for DO (Best *et al.* 2007) versus ASSETS thresholds has an effect on the results of the assessment. We feel the use of the EU-WFD framework in SCB estuaries was well-founded. 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.* unpublished data). Relative to ASSETS, the WFD framework has the advantage of reconciling 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 at full strength seawater but does not take into account effects of salinity (Bricker *et al.* 2003), an issue in estuaries. Thus, applying ASSETS to estuaries with a closed inlet, habitats that are typically brackish and that currently or historically support salmonids in southern California, could be under-protective.

Uncertainties in How Data Are Used to Make an Assessment

This study found that categorization of estuarine eutrophic condition was sensitive to the format of the data as well as the spatial and temporal integration of the data (Nobre *et al.* 2005). Most expert discussion of assessment frameworks tends to focus on the thresholds, with less attention paid to specifying the spatial and temporal density of data and how to use it to make an assessment. Data format and integration were found to impact the condition categories of estuaries for each indicator, regardless of whether the ASSETS or EU-WFD frameworks were applied.

How macroalgal abundance data were used to categorize estuaries had a significant effect on condition category. 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 (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 ($R^2 = 0.691$, $p < 0.0001$, least squares regression, data not shown). Thus, we felt that dry biomass was a more scientifically defensible approach to assessment of eutrophication to eliminate the error involved in variable water content. Using preferred data integration period, eight segments changed category when wet versus dry biomass was used, indicating that this is an important consideration for assessment. Management of spatial data also had an effect on condition categorization. The study utilized average biomass and cover from all three transects to weigh intertidal area in the segment equally and generate a condition class representative of the entire segment rather than the most severely affected subsection. Use of a percentile or only using biomass and cover data from the worst of the three transects generated lower scores in half of the segments, demonstrating the importance of variation in spatial scales in assessment. Finally, how temporal data are integrated also affects how estuaries are categorized. As expected, more segments were categorized as having higher eutrophic condition using annual averages compared to peak season or maximum period; notably, the differences between peak season and the maximum period. For some sites a maximum period with high biomass and cover was averaged with a period of relatively low biomass and cover resulting in a moderate condition category. This approach defined the difference between sites with chronic problems and those with short-duration blooms.

Method of data collection and type of averaging applied to the phytoplankton biomass data set had an impact on the condition categories of a significant number of segments. Half of the systems changed condition category when the discrete data were used versus the continuous data and 25% of the systems crossed the good/moderate boundary indicating a change in whether management action would be taken. This is not surprising, given that phytoplankton biomass in estuaries is highly variable on tidal, weekly and seasonal time scales (Day 1989), so continuous data will always be preferred over discrete grab samples, albeit not always practical. Continuous data could be expressed as either instantaneous 15-minute data or as daily averages; this study used daily averages to eliminate some high frequency noise in the data set. However, comparison between the two data sets indicated that there was not a significant effect on how data were categorized with respect to eutrophic condition.

Data management considerations were also important for determining eutrophic condition based on DO. Both ASSETS and EU-WFD (Bricker *et al.* 2003, Best *et al.* 2007) utilize a percentile approach to data integration, calculated by ranking data from lowest to highest value, and applying the percentile. The EU-WFD applies a 5th percentile and ASSETS a 10th percentile; the 5th percentile of nine months of continuous DO data equates to approximately two weeks below a designated threshold. Use of 5th and 15th percentile relative to the 10th percentiles changes condition classes in 20 to 30% of segments. The use of the percentile approach to integrate duration and frequency of low DO events does not 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. "Natural" hypoxia in bottom waters of bar-built estuaries (Rabalais *et al.* 2010) is potentially an issue for application of DO thresholds and has implications for interpretation this assessment. Shallow estuaries are prone to development of density-driven stratification during restrictions or closure to tidal exchange when the estuaries 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. Studies of natural hypoxia in minimally disturbed "reference" estuaries are needed to clarify this issue.

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