

February 13, 2014

Joint Report of Peer Review Panel

for

**Numeric Nutrient Criteria for the Great Bay Estuary
New Hampshire Department of Environmental Services**

June, 2009

Photo: US Fish & Wildlife Northeast Region



**Victor J. Bierman, Jr., Ph.D., BCEEM
Robert J. Diaz, Ph.D.
W. Judson Kenworthy, Ph.D.
Kenneth H. Reckhow, Ph.D.**

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INTRODUCTION

This peer review was authorized through a collaborative agreement sponsored by the New Hampshire Department of Environmental Services (DES) and the Cities of Dover, Rochester and Portsmouth, New Hampshire. The purpose was to conduct an independent scientific peer review of the document entitled, “Numeric Nutrient Criteria for the Great Bay Estuary,” dated June, 2009 (DES 2009 Report).

The peer review was conducted by a four-person panel (Panel) consisting of:

Victor J. Bierman, Jr., Ph.D., BCEEM
Senior Scientist
LimnoTech
Oak Ridge, North Carolina

Robert J. Diaz, Ph.D.
Professor Emeritus
Virginia Institute of Marine Science
College of William and Mary
Gloucester Point, Virginia

W. Judson Kenworthy, Ph.D.
109 Holly Lane
Beaufort, North Carolina 28516

Kenneth H. Reckhow, Ph.D.
Professor Emeritus
Nicholas School of the Environment
Duke University
Durham, NC

The point of contact for the Panel throughout the review process was Ms. Sally L. Brabble, Litigation Paralegal, at the law firm of Sheehan Phinney Bass + Green in Manchester, New Hampshire.

REVIEW PROCESS

A kick-off teleconference was held on October 29, 2013. Participants on the call included the Panel and representatives from DES and the Cities of Dover, Rochester and Portsmouth. The agenda included a brief history and purpose, an update on contracts, guidance on communications, written public comments, and consideration of a future public meeting. The Panel was given discretion to decide whether to prepare a single consensus report or to submit individual reports.

Subsequent to this kick-off teleconference, requests were made by individual panel members for data, additional data analyses, and additional documents to support their reviews. On November 13, a teleconference was held with DES to discuss these requests and the schedule for DES responses.

On November 18, the Panel requested a time extension from January 31, 2014 to February 28 for completion of its final report. The basis for this request was to allow adequate time for DES to respond to Panel requests for additional information, and for the Panel to review and evaluate this information. On November 25, the sponsors agreed to a compromise time extension to February 19, 2014.

On November 25, an in-person meeting of the Panel was held in Durham, North Carolina. The Panel discussed the DES 2009 Report and the review process. A decision was made to prepare a single joint report, but not to strive for a consensus document. As part of this decision, each panel member agreed to conduct an independent review, arrive at their own independent conclusions, and share written drafts of these conclusions with other members of the Panel.

On November 27, the Panel was provided with numerous written public comments that were solicited as part of the peer review. The Panel was instructed that it was not necessary for them to respond to these comments.

On January 7 and 29, the Panel held two teleconferences to discuss the status of their review comments and the schedule for their final joint report.

PANEL RESPONSES TO CHARGE QUESTIONS

A list of specific charge questions was provided to the Panel for their review of the DES 2009 Report. These charge questions appear below and each is followed by the individual responses of the four panel members. Although this joint report is not explicitly a consensus document, the Panel conclusions reflect high degrees of convergence and agreement.

QUESTION 1. THE REPORT TITLED “NUMERIC NUTRIENT CRITERIA FOR THE . . . GREAT BAY ESTUARY” (hereafter the “DES 2009 REPORT”) was developed over a five-year period starting in 2004. Is the “conceptual model” used in THE DES 2009 REPORT (at 4 and appendix B) to interpret the nutrient criteria reasonably supported by the data and studies for the estuary, the relevant scientific literature and the subsequent information/analyses available for the estuary?

BIERMAN RESPONSE

The “conceptual model” used in the DES 2009 Report is based on the National Oceanic and Atmospheric Administration (NOAA) estuarine eutrophication model (Bricker et al. 2007) that relates external nutrient inputs to primary and secondary symptoms of eutrophication. It also draws upon the conceptual model of coastal eutrophication by Cloern (2001) and technical guidance from the U.S. Environmental Protection Agency (2001) on estuarine and coastal waters.

The “conceptual model” used in the DES 2009 Report is technically sound and widely accepted in the scientific community; however, the site-specific application of this model to Great Bay Estuary for the purpose of developing numeric nutrient criteria did not consider all of the underlying direct/indirect linkages among the relevant stressor variables, response variables and confounding variables. See my response to Question 1e for a more complete discussion.

DIAZ RESPONSE

Much of the conceptual model used to by DES was developed for the first National Estuarine Eutrophication Assessment (NEEA) under the leadership of Suzanne Bricker (Bricker et al. 1999, 2007). The NEEA was a survey of the extent, severity, types, and probable causes of eutrophic symptoms in US coastal systems, with Great Bay being one of 141 systems examined (Figure 1).

The basic NEEA assessment method evaluated eutrophication by examining influencing factors, and primary and secondary symptoms. Overall eutrophic condition and future outlook for eutrophication was then assessed in each system. Bricker et al. (2008) summarized the influencing factors and overall eutrophic condition as:

Great Bay

SUMMARY

In Great Bay, increases in dissolved inorganic nitrogen have occurred over the past 20 years. Increases in chlorophyll a and turbidity have been identified with augmented eutrophication in the inner estuary. As a result, eelgrass biomass has declined by 70% in the last 10 years and the occurrence of nuisance macroalgae is becoming more evident.

Influencing Factors

Any level nitrogen input and low to moderate susceptibility (good ability to dilute and flush nutrients).

Future Outlook

Nutrient related symptoms observed in the estuary are likely to substantially worsen.



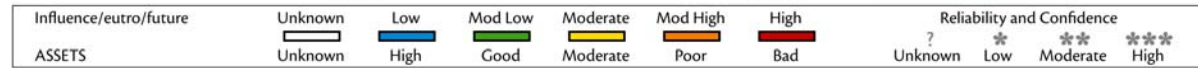
Eutrophic Conditions ***

Primary symptoms high but problems with more serious secondary symptoms still not being expressed.

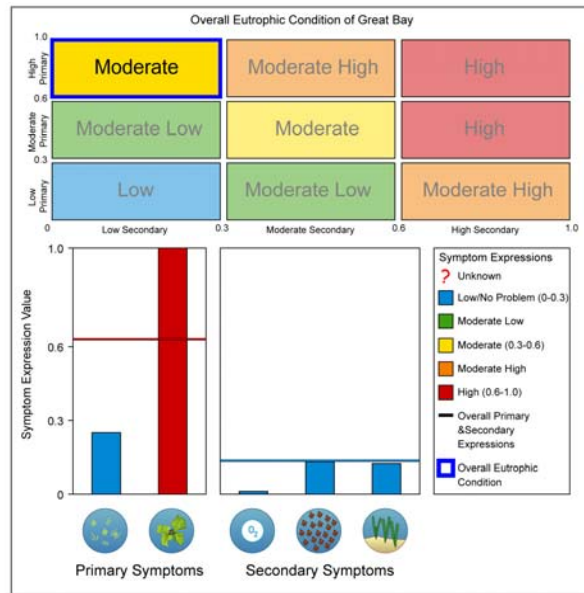
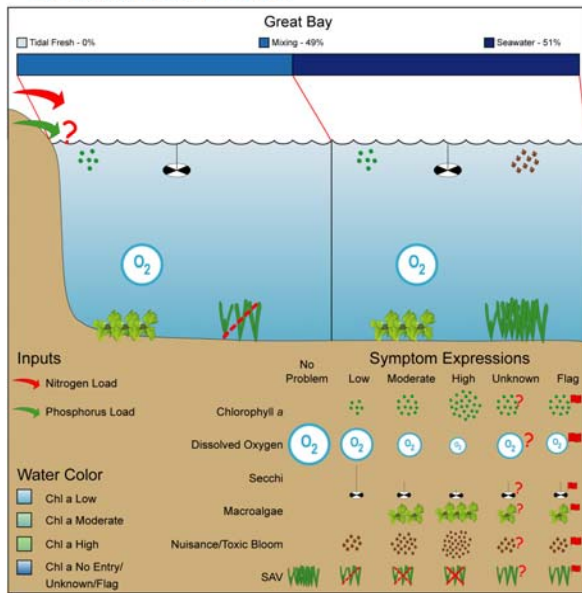


ASSETS Rating

Assessment of Estuarine Trophic Status based on the three factors evaluated in NEEA.



EUTROPHIC CONDITION



WATERSHED AND ESTUARY CHARACTERISTICS

Estuary	Landuse / Population		Watershed Details / Input Loads		
Area (km ²)	47	Urban (km ²)	477 (19.2%)	Area (km ²)	2,555
Tidal fresh zone area (km ²)	0	Agriculture (km ²)	202 (8.1%)	Mean elevation (m)	102
Mixing zone area (km ²)	23	Forest (km ²)	1,740 (70%)	Max. elevation (m)	470
Saltwater zone area (km ²)	24	Wetland (km ²)	65 (2.6%)	Watershed: estuary ratio	54.4
Volume (1,000 x m ³)	177,660	Range (km ²)	3 (0.1%)	TSS (tonne y ⁻¹)	48,800
Depth (m)	3.78	Barren (km ²)	0 (0%)	DIN (kg y ⁻¹)	905,000
Tide Height (m)	2.38	Total (km ²)	2,486 (0%)	DIP (kg y ⁻¹)	Unknown
Residence Time (d)	1	Population	236,203	TSS/est. area (tonne km ⁻² y ⁻¹)	1,038
		Popn: est. area ratio	5,026	DIN/est. area (kg km ⁻² y ⁻¹)	19,255
				DIP/est. area (kg km ⁻² y ⁻¹)	Unknown

Figure 1. Summary of Great Bay from the National Estuarine Eutrophication Assessment update (Bricker et al. 2007)

INFLUENCING FACTORS:

“Influencing factors help establish a link between a system’s natural sensitivity, or susceptibility, to eutrophication as a result of flushing and dilution characteristics, and the nutrient loading and eutrophic symptoms that are observed. In most cases, if the water (and therefore nutrients) is flushed quickly, there is insufficient time for eutrophic symptoms to develop (i.e. low susceptibility). However, if the estuary has a long residence time, there is time for nutrients to be taken up by algae and for blooms to develop. Physical and hydrologic data are used separately to define dilution and flushing ratings which are combined by a matrix to give a susceptibility rating. The susceptibility rating is combined in a matrix with a rating for nitrogen loads to determine the final influencing factor rating. The load component is estimated as the ratio of nitrogen coming from the land (i.e. human-related) to that coming from the ocean (Bricker et al., 2003; Ferreira et al., 2007).”

“The load rating also provides insight into load management since loads that are primarily oceanic in origin will not be easily controlled. In addition to evaluating influencing factors, susceptibility can be used to forecast what symptoms may potentially occur. For example, in some shallow lagoon systems, additional nutrients may result in increased macroalgal abundance rather than high concentrations of phytoplankton/chlorophyll a (Nobre et al., 2005). A typology of these systems is being developed in order to increase assessment and forecasting accuracy by accounting for differences in how systems respond to nutrient inputs (Bricker et al., 2007).”

OVERALL EUTROPHIC CONDITIONS:

“Assessment of the overall eutrophic condition is based on assessment of five symptoms. For each symptom, a level of expression is determined by evaluating the occurrence, spatial coverage and frequency of the symptom. Chlorophyll a and macroalgae are considered primary symptoms that, at high levels, indicate the first stage of water quality degradation associated with eutrophication. The three secondary symptoms represent more serious impacts: low dissolved oxygen levels, loss of submerged aquatic vegetation, and occurrences of HABs. The average of the primary symptoms and the worst case of the secondary symptoms (a precautionary approach) are combined by a matrix to determine the overall eutrophic condition rating.”

“In many estuaries, primary symptoms lead to more serious secondary symptoms. In some cases secondary symptoms can exist in the estuary without originating from primary symptoms. This occurs in many North Atlantic estuaries where some HABs may be transported into the system from the coastal ocean (Bricker et al., 2006). Such systems were consequently given a lower rating (i.e. indicating lesser impact) for HABs than those systems for which these blooms originated within the system. Low ratings, rather than a designation of no problem, were used because it is unclear whether offshore HABs grow and are maintained by landbased nutrient sources once they enter the system.”

“It should be noted that nutrient concentrations are not used as an indicator because they reflect the net biological, physical, and chemical processes such that even a severely degraded system may exhibit low concentrations due to uptake by phytoplankton and macroalgae. Conversely a relatively healthy system might have high concentrations due to low algal uptake, strong filter feeder populations, or may flush nutrients so quickly that phytoplankton do not have the opportunity to bloom extensively.”

The DES 2009 Report did not include sufficient details on Great Bay influencing factors. This becomes an important problem later in addressing many of the charge questions. Of the five symptoms used in the NEEA, the DES 2009 Report used four. Primary symptoms of Chlorophyll a and macroalgae, and secondary symptoms of low dissolved oxygen levels and loss of submerged aquatic vegetation. The occurrence of HABs was not used in the DES 2009 Report because they do not present a threat to Great Bay. The DES 2009 Report added an additional secondary symptom, the effects of accumulated organic matter in sediments on benthic infauna, based on Jim Cloern’s (2001) model of eutrophication impacts. The selection of primary and secondary symptoms was appropriate for assessing eutrophication in Great Bay.

In summary, the Great Bay conceptual model used in the DES 2009 Report included key elements of both the NEEA and Cloern conceptual models but also excluded key elements. The excluded elements were influencing factors (Bricker et al 2008) and complexity of interactions (Cloern 2001) that would have greatly improved interpretation of conditions in Great Bay for setting numerical criteria on nitrogen concentration. DES also used guidance from the 2001 EPA nutrient criteria technical manual (EPA 2001), which has since been reviewed by EPA’s SAB (EPA 2010a), for structuring the case for numeric criteria for nitrogen.

KENWORTHY RESPONSE

The conceptual model utilized by in the DES 2009 Report was based on the NEEA Eutrophication Assessment (NEEA) (Bricker et al. 1999, 2007) and one element of a model developed by Cloern (2001). This approach was an appropriate framework and reasonable starting point to begin the assessment. The NEEA protocol is well supported by the published scientific literature and Cloern’s work is widely cited and recognized as an important contribution to understanding processes of eutrophication. The overall approach calls for assessing five symptoms of eutrophication; two primary symptoms, chlorophyll-a and microalgae, and three secondary symptoms, harmful algal blooms (HABs), low dissolved oxygen levels and loss of submerged aquatic vegetation (eelgrass). DES decided not to include HABs, stating that HABs were not a significant threat to water quality in the Great Bay Estuary. The element of the Cloern model adopted by DES included consideration of sediment organic matter.

In practice, application of the DES conceptual model to the Great Bay Estuary failed to address several influencing factors identified by the NEEA protocol and needed to fully evaluate the effects of nitrogen on eelgrass. Many of the factors explicitly indicated by the NEEA, for example; hydraulic flushing and water residence time (Bricker 1999), were not considered in the DES model. These two physical factors (among several others) are especially important in controlling nitrogen loading, processes of nitrogen

cycling, and nitrogen concentrations in New England Estuaries (Latimer and Rego 2010). Hydrological modelling of individual embayments is a centerpiece of the Massachusetts Estuaries Program (MEP) for developing TMDLs and one of the tools used to evaluate nitrogen loading and its effect on eelgrass. Even though DES cites the MEP work as influential for developing and implementing their approach in the Great Bay estuary, there was no effort made to consider these other important factors.

The NEAA conceptual model was also inadequate because it failed to recognize algal epiphytes as a very important symptom of eutrophication that affects light attenuation and eelgrass abundance (Neckles et al. 1993, Kemp et al. 2004; Ralph et al. 2007). The NEAA and DES did not recognize epiphytes as a contributor to light attenuation; however, empirical research and modelling studies published in the scientific literature clearly demonstrate that one of the primary symptoms of nitrogen over-enrichment and eutrophication in seagrass systems is the overgrowth of micro- and macroalgae on the leaves of seagrasses (Ralph et al. 2007). Much like phytoplankton blooms (water column chlorophyll-a), algal overgrowth on seagrass leaves attenuates light and negatively affects seagrass growth (Neckles et al. 1993, Kemp et al. 2004). Eelgrass beds exposed to eutrophication typically exhibit symptoms which include high epiphyte loading. Additionally, epiphyte loading may be exacerbated by low dissolved oxygen concentrations which limit the metabolism and feeding activities of algal grazers that are primarily responsible for mitigating the potential effects of the excessive growth of epiphytes. Hence, providing a positive feedback mechanism and leading to more excessive epiphyte growth and further light attenuation at the eelgrass leaf surface (Kemp et al. 2004). According to the site specific study referenced in the DES 2009 Report (Morrison et al. 2008), light attenuation in Great Bay can be attributed to water (32%), turbidity (29%), CDOM (27%) and chlorophyll-a (12%). Since these results suggest chlorophyll-a is responsible for only a small fraction of light attenuation and DES implicates nitrogen as the main factor responsible for eelgrass loss, it would be reasonable to evaluate the effect of epiphytes as a diagnostic symptom of eutrophication in the Great Bay system. In their assessment DES did not explicitly state whether they considered epiphytes as a potential eutrophication problem. If we assume that DES did not simply ignore this factor and epiphytes are not contributing significantly to light attenuation, and chlorophyll-a is only a minor contribution to light attenuation, nitrogen cannot be directly implicated as the major cause of light attenuation and eelgrass declines in the Great Bay estuary.

RECKHOW RESPONSE

Yes, the conceptual model is a reasonable choice. Unless stated otherwise, the analyses that I present in this peer review are based on pooling all the stations and sites NH data, which were provided by NH DES.

Specifically respond to the following:

a) Given the available data, is transparency an important factor in the presence/absence of eelgrass in the various segments of the estuary including the upper tidal rivers, great bay, little bay, the Piscataqua River, and/or Portsmouth Harbor? If yes, is it the controlling factor?

BIERMAN RESPONSE

Yes, it is an important factor.

Conceptually, it is the controlling factor, assuming that the prerequisite physical-chemical requirements are met. These include current velocity, waves, tides, salinity, sediment grain size distribution (GSD), sediment total organic carbon (TOC) and sediment sulfide concentration (Koch 2001). Another important factor is explicit consideration of estuarine bathymetry. The data and analyses in the DES 2009 Report did not adequately demonstrate that transparency is the controlling factor in Great Bay Estuary because it did not explicitly investigate any of these confounding factors.

DIAZ RESPONSE

Yes, transparency is one of many important factors that affect eelgrass presence/absence and growth/health.

KENWORTHY RESPONSE

Water transparency controls the availability of light for underwater plants and is an important factor controlling the distribution and abundance of seagrasses, including eelgrass (Dennison and Alberte 1982, 1985, 1986, Dennison et al. 1993). When considering all of the potential symptoms of eutrophication and the stressor response relationships, the most well documented, consistent, and empirically supported relationship is between transparency and eelgrass response (Krause-Jensen et al. 2008). DES was correct in considering transparency as a factor in their assessment. But transparency is not the only important factor, and the relationship of eelgrass loss to transparency varies by estuarine water depth with the greatest eutrophication induced responses documented for deeper waters. The effect of transparency will also be influenced by the light requirements of eelgrass which may be different depending on ambient water quality and sediment conditions (Kenworthy and Fonseca 1996, Duarte et al. 2007, Kenworthy et al. 2013). Eelgrass growth, abundance and distribution are also controlled by temperature, nutrient availability (primarily nitrogen and phosphorus), tidal range, water motion, wave action, water residence time, bathymetry, substrate type, substrate quality, severe storms, disease, plant reproduction and anthropogenic disturbances (Short and Wyllie Echeverria 1996, Koch 2001, Short et al. 2002, Kemp et al. 2004, Moore and Short 2006, Krause-Jensen et al. 2008). Eelgrass distribution and abundance in an estuary results from the complex interaction of some or all of the factors listed above, and no two estuaries or sub-embayments of an estuary are identical in all of these factors (see Figure 5 in Krause-Jensen et al. 2008). In order to determine if one or more of these are “controlling” it would be necessary

to either consider all of them and their interactions, or be able definitively eliminate certain factors as insignificant contributors.

In their assessment of transparency and its controlling effect on eelgrass, DES considered a sub-set of the factors listed above; nutrient availability, tidal range, disease, and two anthropogenic factors (dredging and mooring fields) (R-WD-09-12, Numeric Nutrient Criteria for the Great Bay estuary). Because nutrient availability does not directly affect transparency, this factor was addressed indirectly through the interaction between nitrogen concentration and chlorophyll-a using a linear regression approach between chlorophyll-a and the light attenuation coefficient measured as K_d . Tidal range was incorporated into the assessment using a model developed by Koch (Koch and Beer 1996, Koch 2001). Disease was incorporated into the assessment of eelgrass status in the Great Bay Estuary by considering the effects of wasting disease on temporal and spatial fluctuations in eelgrass cover (WD Doc R-WD-08-18, Methodology and Assessment Results related to Eelgrass and Nitrogen in the Great Bay Estuary for Compliance with Water Quality Standard for the New Hampshire 2008 Section 303(d) List). The eelgrass assessment also considered historical and current information on the potential effects of dredging and vessel mooring fields on eelgrass. A critical deficiency in the DES 2009 Report was the fact that DES did not attempt to present evidence for ruling out the other factors listed above that could be controlling the presence or absence of eelgrass (e.g., temperature, water motion, wave action, bathymetry, water residence time, substrate type, substrate quality, severe storms, disease, epiphytes, and plant reproduction). Before critically evaluating the DES approach and responding to the remaining questions, it is first necessary to consider how DES evaluated the status of eelgrass in the Great Bay Estuary.

DES 2008 EELGRASS ASSESSMENT METHODS USED FOR THE 2009 REPORT AND ESTABLISHMENT OF NITROGEN CRITERIA FOR THE GREAT BAY ESTUARY

As per the New Hampshire Department of Environmental Services (DES) assessment methodology used in the DES 2008 and 2009 Reports, a significant loss of eelgrass (*Zostera marina*) habitat would constitute a violation of the narrative standard ENV-Ws 1703.19 and create a water quality standard violation for biological integrity (WD Doc R-WD-08-18). To conduct the assessments, DES originally selected eelgrass cover data in 11 geographically distinct assessment zones as an indicator for water quality impairment determinations (see Table 2 in WD Doc R-WD-08-18). The 11 original zones were assessed for eelgrass cover up to 2005 and have since been subdivided into 13 zones by DES after recognizing one further subdivision of the Piscataqua River and distinguishing Portsmouth and Little Harbors as distinct zones (Table 1). DES eelgrass cover data are now available up to 2012, but the original assessment terminated in 2005. The historical cover data for the DES 2009 Report were derived from a wide range of sources dating back to 1948 and considered the catastrophic effects of the wasting disease on eelgrass in 1931-1932, and subsequent wasting disease events in 1984 and 1988-1989. Standardized mapping of eelgrass cover in the Great Bay Estuary using nationally accepted protocols was not initiated until 1986 and included only the assessment zones in Great Bay, and the Winnicut, Squamscott and Lamprey Rivers. All 13 zones were first simultaneously mapped with standardized protocols for eelgrass cover in 1996 and again in 1999 with a continuous annual record that is now available between 1999 and 2012.

To conduct their assessments of changes in eelgrass cover in each of the zones DES used two methods; a zone will be considered to have significant eelgrass loss if 1) the change from historic levels is >20% or, 2) where annual surveys were available, there was a statistically significant ($p < 0.05\%$) decreasing trend that shows a loss of 20%. To further minimize the effect of annual variation on the analyses conducted with method #1, DES used the median eelgrass cover for three years (2003, 2004, and 2005) as the contemporary reference point to compare with the historical data. Selection of the 20 % threshold was based on an analysis of the variation in a continuous record of eelgrass cover (1990-1999) in one zone, Great Bay. This analysis indicated a relative standard deviation of 6.5%. DES selected three relative standard deviations as the threshold for the upper bound of natural variation ($3 * 6.5$) to constitute the threshold and assumed that the variation in cover in Great Bay was representative of the other zones in the Estuary. Currently there is no citable published precedent in the scientific literature for using a threshold of 20% change in any eelgrass monitoring program, and the implications for selecting a threshold of 20% change could be very different depending on the time interval over which the change occurs. To the best of my knowledge, the State of Washington conducts the only long-term and large-scale statewide eelgrass monitoring program in the country that utilizes a threshold change criteria of 20%. The Washington State eelgrass sampling program is probabilistic based, and the criteria are time dependent, utilizing data from annual monitoring of eelgrass cover over a period of 10 years to conduct the change analysis. It is also my understanding that the Washington State monitoring program is undergoing an internal technical and quality control review. The DES eelgrass monitoring program could gain substantial benefits and a better understanding of the difficulties in establishing statistically significant eelgrass change detection by consulting the Washington State program.

DES EELGRASS ASSESSMENT RESULTS AND NITROGEN CRITERIA

Seven of the original 11 zones (Squamscott, Lamprey, Oyster, Belamy, Little Bay, Upper Piscataqua and Lower Piscataqua) were assessed using method #1, based on historical data from 1948, 1962 and 1980-81. All seven of these assessments met the DES criteria for impairment with respect to eelgrass status. In two zones, Upper and Lower Piscataqua Rivers, different years were combined to get historical reference totals, but the DES 2009 Report does not provide adequate explanations justifying the combination of data collected 20 years apart.

Four of the original 11 zones (Winnicut, Great Bay, Portsmouth Harbor/Little Harbor and Sagamore) were assessed using method #2, based on linear regressions. Winnicut River and Great Bay were analyzed for the period 1990-2005, and Portsmouth Harbor/Little Harbor and Sagamore Creek for the period 1996-2005. Of these four zones, the only significant decreasing trend was detected in the Winnicut River. In Great Bay, where the largest amount of eelgrass resource is found in the estuary, eelgrass actually increased from historical levels reported in both 1948 and 1980-1981 and the median cover for years 2003-2005 was higher than the cover in 1990 at the start of the regression analysis.

CRITICAL EVALUATION OF THE DES EELGRASS ASSESSMENT METHOD

The DES assessment reports were correct in dividing the Great Bay Estuary into 11 distinct assessment zones, each with independent analyses of eelgrass status. This approach is consistent with the approach taken by the Massachusetts Estuaries Project in assessing nitrogen loading in coastal embayments. A

main strength in this approach is that it implicitly recognizes heterogeneity in the estuarine system as well as the possibility that there may be important differences in the biophysical characteristics of the zones which could affect eelgrass distribution and abundance. Spatial variation in factors such as natural watershed landscape characteristics, non-point source water runoff, water depth, sediment type, substrate stability, wind and wave exposure, tidal velocities, freshwater discharge, non-point source runoff, groundwater discharge and land use are known to interact and determine different eelgrass distributions in shallow water coastal ecosystems (Thayer et al. 1984, Larkum et al. 2006, Orth et al. 2010a, b). Stochastic events like severe storms, ice scour and climate variation were not considered even though these are known to affect eelgrass (Frederiksen et al. 2000 a, b, Orth and Moore 2006, Krause-Jensen et al. 2008). The assessment completely ignored biological aspects of the system including plant reproduction, grazing and bioturbation. Some these factors can limit eelgrass growth, reproduction and distribution to the extent that the species can be completely eliminated from an estuary (see Figure 5 in Krause-Jensen et al. 2008). The importance of considering multiple controlling factors also directly applies to eelgrass restoration as empirically determined by Short et al. (2002) in Great Bay and elsewhere in New England estuaries. These confounding factors can obscure the relationship between nitrogen loading/eutrophication and eelgrass response, therefore the assessment was weakened by not explicitly considering any of these factors in their evaluation of eelgrass loss. The DES case was further weakened by only considering information for the anthropogenic effects of dredging and the existence of mooring fields as other potential factors controlling eelgrass distribution in the Great Bay estuary.

DES was correct to factor in consideration of the effects of “wasting disease” on eelgrass populations and the general distribution and abundance of eelgrass in the Great Bay Estuary, but the effects of wasting disease on eelgrass should have been considered uniformly across all of the different zones. In five zones which utilized historical data as a baseline from 1948, 1962, and 1980-81 (Oyster, Bellamy, Little Bay, Upper Piscataqua, Lower Piscataqua), DES failed to acknowledge the possible effects of wasting disease events that were reported in the estuary in 1984 and 1989. Reliable and consistent mapping data for these five zones was not available until 1996, so it was incorrect to identify the baseline cover prior to the two contemporary wasting disease events if their effect could not be assessed. A further indication that DES was inconsistent in their consideration of the wasting disease was the fact that DES explicitly recognized the effects of the disease in the Great Bay zone and did not initiate the regression analysis for the assessment until 1991.

RECKHOW RESPONSE

Transparency is one of several factors determining the presence/absence of eelgrass. It is no more/less controlling than is TN. To see this Kd-TN covariation conundrum, consider the graph below:

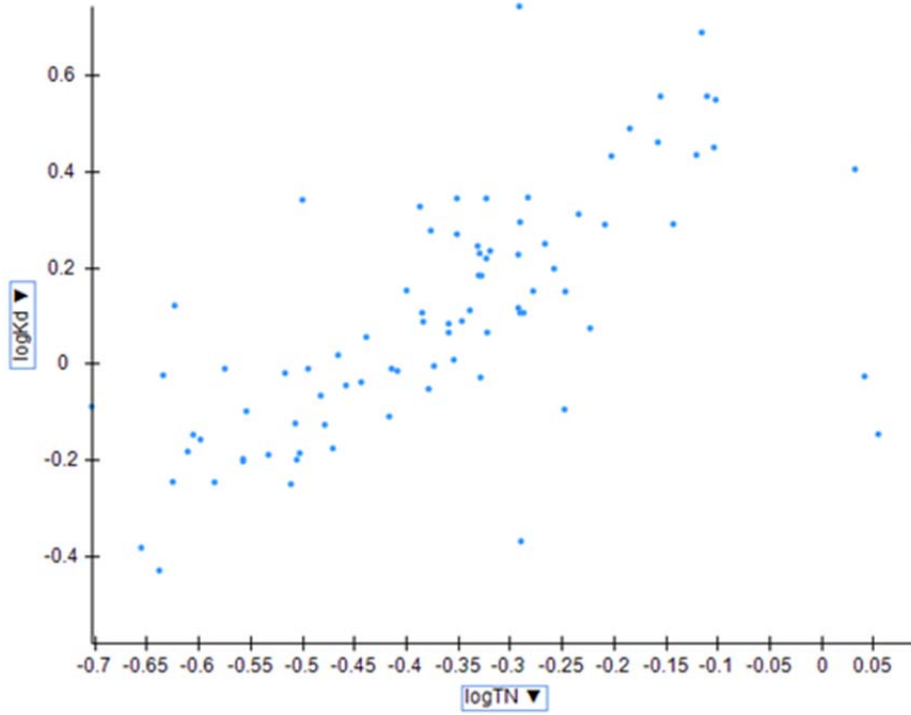


Figure 2. Log Kd versus Log TN

TN and Kd are highly correlated, which is likely due to the fact that TN affects algae (chlorophyll), and algae affects Kd. Based on scientific knowledge, this is a plausible causal relationship, and it is supported by this graph.

b) Given the available data/studies, is nitrogen an important factor in the presence/absence of eelgrass in various segments of the estuary (please be specific in terms of the impact or lack of impact in the tidal rivers, great bay, little bay, the Piscataqua River, or Portsmouth Harbor)? If yes, is it the primary factor?

BIERMAN RESPONSE

Yes, it is an important factor.

It is one of the primary factors, not the sole primary factor. Light is also a primary factor. Other important factors are those listed above in my response to Question 1a. The DES 2009 Report did not adequately demonstrate that nitrogen is the primary factor in the Great Bay Estuary because it did not explicitly consider any of the other important, confounding factors in developing relationships between nitrogen and the presence/health of eelgrass.

These answers apply to the Estuary as a whole and to its various individual segments.

DIAZ RESPONSE

Yes, overall nitrogen is an important factor for eelgrass growth, but in the context of numeric nitrogen criteria it is the concentration of nitrogen that disrupts the balance of primary producer species that are known to negatively interact with eelgrass (Neckles et al. 1993). With increasing nutrients there is a shift in primary producers from perennial macroalgae and seagrasses toward a dominance of ephemeral macroalgae, epiphytes and phytoplankton (Neckles et al. 1993, Cloern 2001). Within the various estuarine segments, the importance of nitrogen as a controlling factors needs to be balanced by other co-varying factors, such as transparency and sediment quality (Kenworthy et al. 2013), and those listed in Kenworthy's response.

KENWORTHY RESPONSE

The eelgrass assessment document (WD Doc R-WD-08-18) states explicitly that DES used a methodology to determine compliance with water quality standards for biological integrity using eelgrass cover as an indicator. In this document DES also included their assessments of chlorophyll-a in each of the zones and determined that there were four zones with nitrogen impairment and seven zones without nitrogen impairment; implicitly linking eelgrass impairment to nitrogen impairment. Four of the seven zones with eelgrass impairment were not declared nitrogen impaired. This is not very compelling evidence linking nitrogen impairment to eelgrass impairment if only 36% of the zones in the Great Bay Estuary are considered impaired for both, and more than half of the zones with eelgrass impairment were not declared nitrogen impaired. Even if it were correct to infer a linkage between chlorophyll-a data, nitrogen impairment and eelgrass status in the Great Bay estuary, it was inappropriate for the DES to imply a linkage in the 2008 and 2009 reports. The chlorophyll-a data were from the period 2000 – 2008 while the eelgrass changes in the zones that were declared impaired are based on historical data from 1948, 1962 and 1980-81 and changes that may have occurred during extended periods without eelgrass

cover data or environmental monitoring. There is no basis for a scientifically defensible linkage between nitrogen impairment and eelgrass impairment presented in the report.

DES eelgrass cover data using standardized protocols is now available up to 2012 with a nearly spatially complete and continuous 17 year record beginning in 1996 (Table 1). The potential implications of more recent eelgrass cover data were suggested in the DES 2009 Report on page 66 where DES acknowledges that even where the lowest concentrations of nitrogen occur in Portsmouth and Little Harbors conditions are not pristine and eelgrass is declining. There are several aspects of this more recent data set that provide significant insights and improvements in the assessment of eelgrass status with regard to nitrogen impairment and other factors that may affect eelgrass growth and distribution. First of all, the more recent 17 year record avoids the uncertainties associated with unreliable and aggregated historical data sets used in the original assessment and eliminates the necessity of using two different assessment methods. It would also avoid uncertainties and assumptions about undocumented effects of wasting disease and other environmental stressors that may have affected eelgrass during periods of time when environmental monitoring was incomplete and eelgrass cover was not assessed between 1948 and 1996. Second, the more contemporary eelgrass data set would coincide more closely with regularly monitored environmental data (e.g., chlorophyll a, light attenuation, salinity, temperature, etc.) in the estuary presented in the 2009 DES Report.

A preliminary analysis of this more recent eelgrass cover data set re-affirms the DES general concerns for the declining status of eelgrass cover throughout the Great Bay estuary (Table 1). With the exception of just one zone, Little Bay, all of the individual zones with eelgrass cover present in 1996 displayed >20% declines and should the linear regressions be significant as per the DES criterion #2, these zones could be legitimately declared impaired with regard to eelgrass cover. Furthermore, this preliminary analysis suggests that the Great Bay zone (- 35.95% decline) could be considered impaired, whereas in the original assessment it was ambiguously declared threatened. Little Bay actually displayed a small increase (5.81%). Three zones could not be assessed because there was no eelgrass cover in 1996. In two of these zones (Lamprey and Bellamy Rivers) small coverage's of eelgrass were detected during the time interval, but eelgrass was not present in 2012.

In addition to this preliminary assessment of eelgrass status, I have also calculated specific rates of annual change in the different zones of the Great Bay estuary ($\% y^{-1}$; see Table 1) and compared these to the reported rates of decline for seagrasses in general (Waycott et al. 2009) and eelgrass in Massachusetts (Costello and Kenworthy 2011). Rates of decline ranged from - 2.43 $\% y^{-1}$ in Sagamore Creek to - 6.25 $\% y^{-1}$ in the upper Piscataqua River. The average rate of decline for the nine zones that could be analyzed was - 4.19 $\% y^{-1}$. Based on a global assessment of 217 sites, the median annual rate of decline for seagrasses since 1980 was - 5 $\% y^{-1}$ (Waycott et al. 2009). This global assessment attributed seagrass declines to overexploitation, physical modification, nutrient and sediment pollution, introduction of nonnative species, and global climate change. Nearby in Massachusetts, where there has been well documented concern for nutrient impairments derived from groundwater nitrogen discharges, a statewide change analysis of eelgrass abundance over a period of 12 years reported rates of decline ranging from - 2.21 $\% y^{-1}$ to -3.51 $\% y^{-1}$. In some of the most nutrient impaired embayments in Massachusetts eelgrass

has been completely extirpated in the past three decades. Based on the average rate of decline in the Great Bay estuary, eelgrass could be lost from large portions of the system in two to three decades.

As suggested above, the preliminary analysis using the more current eelgrass cover data affirms scientifically defensible DES concerns for eelgrass declines in the Great Bay estuary; however, by no means does this infer a direct relationship with nitrogen impairment as suggested by the original assessment in WD Doc R-WD-08-18, Methodology and Assessment Results related to Eelgrass and Nitrogen in the Great Bay Estuary for Compliance with Water Quality Standard for the New Hampshire 2008 Section 303(d) List. In fact, this new analysis confirms a fundamental flaw in the DES approach to setting nitrogen concentration criteria using the regression method in the DES 2009 Report. This DES 2009 Report clearly shows that nitrogen (Figures 4, 6, and 17), chlorophyll-a (Figures 13, 14 and 17) concentrations, light attenuation (Table 9) and turbidity (Figure 17) are highest in the tidal tributaries and are progressively diluted by ocean water down to the mouth of the estuary. Yet, even at locations furthest downstream from the tributaries (e.g., Portsmouth Harbor, Little Harbor and the Lower Piscataqua River) with the lowest concentrations of chlorophyll-a, nitrogen, turbidity, and greater water transparency, eelgrass is declining at significant rates (Table 1). Two alternative conclusions can be drawn from this analysis; 1) either the proposed nitrogen criteria will not protect eelgrass from further declines or, 2) there are other factors contributing to the eelgrass fluctuations and declines which have not been adequately addressed in the DES assessment reports.

Table 1. Annual estimates of eelgrass cover and percent change in cover (total change and percent per year) for the Great Bay Estuary between 1996 and 2012.

Data provided on my request by the New Hampshire Department of Environmental Services.

nd = no data collected, na = data not applicable

YEAR	Winnicut River	Squamsco tt River	Lamprey River	Oyster River	Bellamy River	Great Bay	Little Bay	Upper Piscataqua River	Lower Piscataqua River North	Lower Piscataqua River South	Portsmouth Harbor	Little Harbor	Sagamore Creek
1996	7.6	0	0	14	0	2495.4	32.7	1.6	20.9	10.2	245.6	70.1	1.8
1997	7.5	0	0	nd	0	2297.8	Nd	Nd	Nd	nd	Nd	Nd	Nd
1998	10	0	0	nd	0	2387.8	Nd	Nd	Nd	nd	Nd	Nd	Nd
1999	10.2	0	0	0	0	2119.5	26.2	0.5	7.4	4	244	50.1	3
2000	0	0	0	0	0	1944.5	7.5	1.6	3.8	7.6	260.5	60.9	0.9
2001	4.1	0	0	0	0	2388.2	10.9	2	9.7	10.7	274.2	45.3	2.2
2002	3.5	0	0	0	0	1791.8	4.3	0.5	8	9.3	268.9	63.1	2.3
2003	3.5	0	2.2	0	0	1620.9	14.2	2.9	22.9	9.2	270.1	54.7	2.2
2004	4.2	0	0	0	0.8	2037.6	12.8	0.7	13.5	6.5	225.2	65.8	2.5
2005	9.1	0	0	0	0	2165.7	25.8	0.4	14.5	9.6	232.5	47.9	6.1
2006	0.8	0	0	0	0	1319.8	12.2	0.8	10.8	11.6	217.6	52.1	0.9
2007	0	0	0	0	0	1245.3	0.1	0	0.4	5.6	201.3	42.7	0.6
2008	0	0	0	0	0	1394.9	0	0	0	3.9	183.8	41.4	2.3
2009	0.1	0	0	0	0	1700.6	0	0	0	6.4	155	30.2	0.5
2010	0	0	0	0	0	1722.2	0.3	0	0	3.5	128	42.5	0.2
2011	0	0	0.5	0	0	1623.2	48.2	0	0	6.9	178.8	31.6	1.5
2012	0.3	0	0	0	0	1598.4	34.6	0	1.6	5.1	139.6	36.4	1.1
Percent Change	-96.05	na	na	-100.00	-100.00	-35.95	5.81	-100.00	-92.34	-50.00	-43.16	-48.07	-38.89
Percent Change Per Year	-6.00	na	na	-6.25	na	-2.25	0.36	-6.25	-5.77	-3.13	-2.70	-3.00	-2.43

RECKHOW RESPONSE

To assess the importance of several factors on eelgrass areal coverage, I applied some multivariate pattern recognition techniques to the NH data. One approach that I used is called a “classification tree.” A classification tree is somewhat like a multiple regression model, in that it yields a predictive model for a single variable. The response variable in regression is usually continuous, while the response variable in a classification tree is categorical. Even though eelgrass areal coverage is a continuous variable, I chose to apply a classification tree (with five discrete classes for eelgrass acreage) because the tree diagram provides an informative visual display.

Category	Eelgrass Acreage
1	0
2	0 < eelgrass < 10
3	10 < eelgrass < 70
4	120 < eelgrass < 275
5	eelgrass > 1200

Figure 3. Categories for Eelgrass

A classification tree partitions the multidimensional space of the predictor variables into regions, each of which is assigned to a category for the response variable. One way to fit classification tree models and partition the space of predictor variables into classes can be described on a conceptual level as follows. At the beginning, all observations are signed to the same class (or given the same prediction for a regression tree). In the first step the observations are split into two classes based on a criterion that minimizes the impurity or maximizes the homogeneity of each class. Additional splits are determined on a step-by-step basis, not optimized for the entire tree at one time. At each split, the predictor variable that minimizes a classification error criterion is selected to provide the classification split.

The next figure presents the best-fit model using the predictor variables chlorophyll, salinity, SS, TN, TP; the five eelgrass categories for the response variable are presented in the table. Note that salinity is the first (most important) split/classification predictor variable. Other variables provide optimal splits down in the branches of the tree. The interpretation of this classification tree is as follows. Applying the classification tree algorithm, we find that salinity is the best classifier for the five eelgrass categories, so starting at the top of the tree, if salinity is less than 26.55 (the number in the top circle) for a data point, we move to the left branch. If salinity is greater than 26.55 we move to the right.

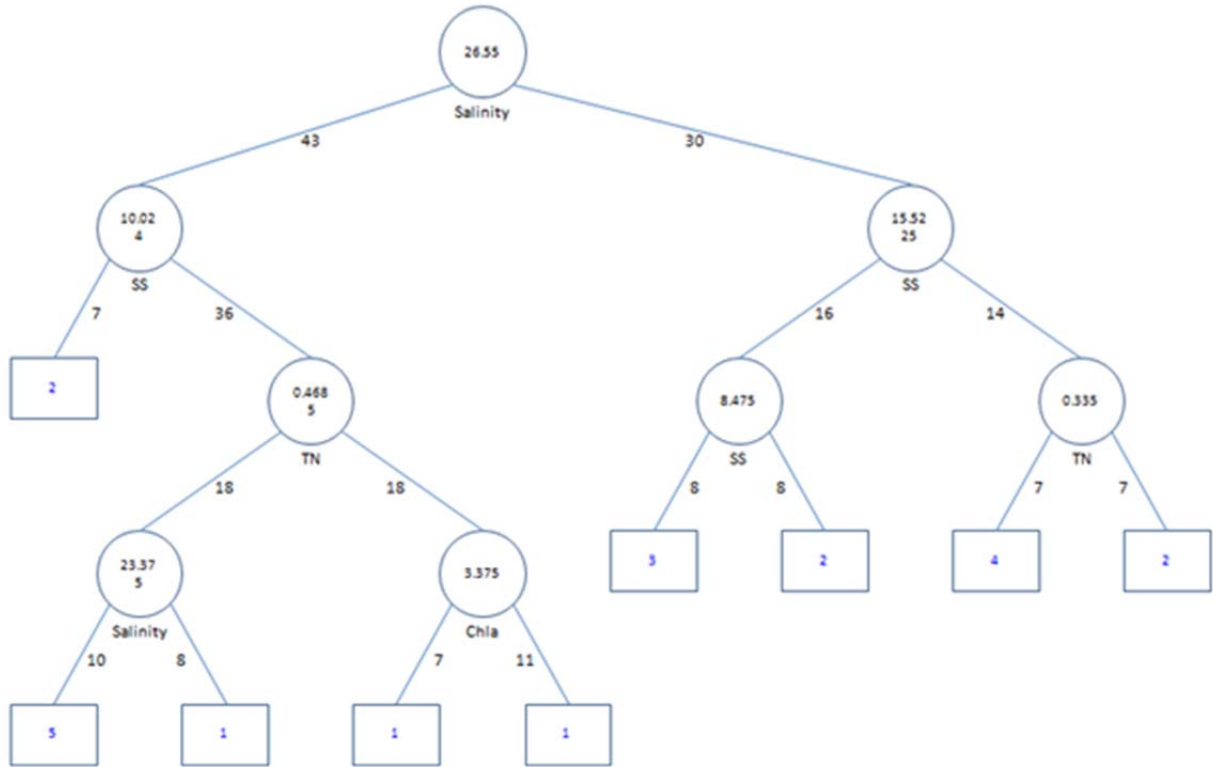


Figure 4. A “best fit” Classification Tree

Moving down the left branch, the next split for a classifying variable is for SS (suspended solids); in this case if the suspended solids concentration is less than 10.024, we go down the left branch, which leads to the data point being placed in eelgrass category two ($0 < \text{eelgrass} < 10$). If the suspended solids concentration is greater than 10.024, we move down to the right where the next classification split is for TN.

This classification tree, and other analyses I have undertaken on the NH data set, clearly show the multivariate relationships among the variables. The effectiveness of TN load reductions clearly varies from site to site, suggesting that a site-specific numeric nutrient criterion should be considered.

c) Does the available information indicate that increased algal growth is causing or significantly contributing to a loss of eelgrass and that nitrogen reductions will significantly improve the conditions for eelgrass growth and/or restoration?

BIERMAN RESPONSE

According to Piscataqua Region Estuaries Partnership (PREP) (2013), phytoplankton chlorophyll-a concentration has not shown a positive or negative trend in Great Bay between 1975 and 2011. The NOAA national eutrophication assessment covering the 1990s and early 2000s (Bricker et al. 2007) also indicated that chlorophyll-a was not a serious problem, and may have declined over the assessment period.

No data were presented for algal growth (primary productivity) in either the PREP (2013) report or the DES 2009 Report. In both of these reports, phytoplankton are characterized exclusively by chlorophyll-a concentration, a snapshot-in-time measurement that represents standing stock, not rate of algal growth.

As stated above in my response to Question 1b, light is one of the primary factors controlling eelgrass growth and, presumably, restoration. Conceptually, if nitrogen concentration and chlorophyll-a were positively correlated, then reductions in nitrogen could lead to improvement in underwater light because chlorophyll-a is one of the factors contributing to underwater light attenuation.

In the Great Bay Estuary, the following are the contributions to K_d (underwater light attenuation coefficient) from the site-specific model by Morrison et al. (2008): water (32%), turbidity (29%), CDOM (27%) and chlorophyll-a (12%). The DES 2009 Report assumes that CDOM is not related to algal primary productivity and ignores water because it is a constant background component.

An immediate observation is that not only is chlorophyll-a a small component of K_d , median chlorophyll-a concentrations in Great Bay are low and range between 1-7 $\mu\text{g/l}$ (Table 6). It is unlikely that reductions in nitrogen concentration could cause significant improvements in light by causing reductions in chlorophyll-a concentration.

To establish a scientifically defensible linkage between nitrogen concentration and K_d in Great Bay Estuary, the following multiple, component linkages require investigation using more than a simple linear regression (SLR) analysis:

1. Chlorophyll-a versus nitrogen
2. POC versus chlorophyll-a
3. K_d versus POC
4. K_d versus turbidity
5. Turbidity versus POC
6. Turbidity versus ISS
7. K_d versus chlorophyll-a

The first three component linkages represent the conceptual model for the influence of nitrogen concentration on Kd (Benson et al. 2013). That is, $N \Rightarrow \text{chlorophyll-a} \Rightarrow \text{POC} \Rightarrow \text{Kd}$. The fourth component linkage is important because the model by Morrison et al. (2008) indicates that turbidity makes a significant contribution to Kd in Great Bay Estuary. The fifth and sixth linkages are important because the DES 2009 Report states on Page 64 that turbidity in Great Bay Estuary is due to POC and ISS. The seventh linkage is important because it must be shown that changes in chlorophyll-a concentration (due to changes in nitrogen concentration) can cause significant changes in Kd.

The DES 2009 Report showed SLR results for chlorophyll-a versus nitrogen concentration (Figures 15 and 17), but not for POC versus chlorophyll-a or Kd versus POC. It showed SLR results for turbidity versus POC (Figure 35) but not for Kd versus turbidity, turbidity versus ISS or Kd versus chlorophyll-a.

Results in Figure 35 indicate that POC explains 47% of the variability in turbidity. Consequently, POC explains 47% of the variability in the factor that accounts for 29% of the contribution to Kd. Total dissolved nitrogen (TDN) explains 34%/46% (maximum/minimum) of the variability in POC (Figure 34). Consequently, TDN explains less than half of the variability in POC, which itself explains less than half of the variability in the factor that accounts for 29% of the contribution to Kd.

Regarding the SLR results for chlorophyll-a versus nitrogen concentration (Figures 15 and 17), DES provided, at my request, supplementary SLR results with nitrogen concentration replaced by salinity for Figure 15. The strength of the correlation with salinity was comparable to that with nitrogen concentration. Given the strength of the correlation between salinity and nitrogen concentration (Figure 21; $R^2 = 0.680$), this makes sense. Consequently, the results in Figure 15, in conjunction with the supplementary results with nitrogen replaced by salinity, demonstrate only that chlorophyll-a and nitrogen concentrations co-vary with salinity, not that there is a causal relationship between chlorophyll-a and nitrogen.

Results in Figures 36-37 for regressions of turbidity versus nitrogen concentration appear spurious because there was no explicit consideration of the co-varying/confounding factors ISS or salinity. Similarly, results in Figures 38-39 for regressions of Kd versus nitrogen concentration appear spurious because there was no explicit consideration of the co-varying/confounding factors ISS or salinity. In fact, when salinity is substituted for nitrogen concentration as the independent variable in Figure 38 (supplementary results provided by DES at my request), the correlation between Kd and salinity is almost as strong as the correlation between Kd and nitrogen concentration. Consequently, the results in Figure 38, in conjunction with the supplementary results with nitrogen replaced by salinity, demonstrate only that Kd and nitrogen concentration co-vary with salinity, not that there is a causal relationship between Kd and nitrogen.

In summary, the DES 2009 Report failed to analyze all of the linkages (co-varying factors) between nitrogen concentration and Kd in their conceptual model of Great Bay Estuary. My opinion is that the SLR results in Figures 38-39 for Kd versus nitrogen concentration are based on weak evidence and are unreliable due to lack of explicit consideration of all the underlying direct/indirect linkages among the relevant stressor variables, response variables and confounding variables.

DIAZ RESPONSE

While declining seagrass in favor of macroalgae or phytoplankton is an all too common response from nutrient driven eutrophication (Burkholder et al. 2007), a causal link between eelgrass and macroalgae in Great Bay is not clear. Monitoring data and other published accounts indicate the occurrence of macroalgae has increased in Great Bay, but there does not seem to be any strong association between macroalgae and eelgrass in Great Bay. Relative to nutrients, Burkholder et al. (2007) say the following in their abstract:

“The most common mechanism invoked or demonstrated for seagrass decline under nutrient over-enrichment is light reduction through stimulation of high-biomass algal overgrowth as epiphytes and macroalgae in shallow coastal areas, and as phytoplankton in deeper coastal waters. Direct physiological responses such as ammonium toxicity and water-column nitrate inhibition through internal carbon limitation may also contribute. Seagrass decline under nutrient enrichment appears to involve indirect and feedback mechanisms, and is manifested as sudden shifts in seagrass abundance rather than continuous, gradual changes in parallel with rates of increased nutrient additions.”

At some level, macroalgae, eelgrass, and nitrogen are connected and interact. The DES 2009 Report is a step in the right direction, but the approach taken needs to include the complexity of interactions as pointed out by Burkholder et al. (2007) and Kenworthy et al. (2013), and the other peer review responses to this question.

KENWORTHY RESPONSE

Strictly speaking this question is difficult to answer because there are no direct measurements of algal growth in the DES 2009 Report. All data for algae in the water column are presented as concentrations of chlorophyll-a, not phytoplankton growth. Macroalgal growth was considered in their assessment as stated explicitly in the 2009 DES Report *“The nitrogen threshold for the protection of eelgrass was derived using a weight of evidence approach which included the thresholds for macroalgae proliferation”*, assuming proliferation implies growth. In a scientific context ‘proliferation’ is an ambiguous term and DES should have used a more direct and quantitative metric for characterizing macroalgae. However, it was clear that the DES 2009 Report attempted to attribute eelgrass loss to the growth of macroalgae and incorporate this information into the weight of evidence for determining nitrogen thresholds for eelgrass.

The DES was correct in their consideration of the potential effects of macroalgae on seagrass distribution and abundance in the Great Bay estuary and as possible evidence for nitrogen impairment. It is well documented that the proliferation of ephemeral macroalgae (e.g., *Gracilaria* and *Cladophora spp*) from nitrogen enrichment can negatively affect the distribution and abundance of seagrasses in general (McGlathery et al. 2007), and eelgrass in particular (Valiela et al. 1992, Short and Burdick, 1996, Hauxwell et al. 2001, Hauxwell et al., 2003). There is compelling scientific evidence that if nitrogen levels exceed seagrass nutrient requirements macroalgae can proliferate to the extent that it will shade eelgrass and outcompete eelgrass for light. Blooms of macroalgae can displace eelgrass and affect the

physical and chemical properties of the sediment to the extent that the blooms will limit distribution and abundance or prevent the re-growth of eelgrass.

As weight of evidence for determining nitrogen thresholds based on the status of macroalgae in the Great Bay estuary, DES was unable to definitively document spatial or temporal trends in macroalgae distribution and abundance for the period during which eelgrass declines were documented. The DES 2009 Report cited historical studies of macroalgae species composition and distribution as well as references to anecdotal reports of increases in nuisance macroalgae, but made no attempt to quantitatively summarize the baseline of macroalgae species composition and distribution or temporal changes in macroalgae abundance that coincided with most of the eelgrass declines (see page 37 in R-WD-09-12, Numeric Nutrient Criteria for the Great Bay Estuary). Hence, any relationship between nitrogen impairment, macroalgae growth and eelgrass abundance cannot be supported.

In an attempt to establish a relationship between total nitrogen concentration and the potential effects of macroalgae on eelgrass in the Great Bay estuary, DES relied on a single study conducted in 2007 in the Great Bay zone only (Pe'eri et al. 2008). This remote sensing study utilized hyperspectral imagery to map eelgrass beds and macroalgae mats in Great Bay and relied on another monitoring survey (Short, 2008) to ground truth and verify the benthic habitat signatures. Using the information from these two studies the DES 2009 Report (see page 37) identified 1246 acres of eelgrass and 137 acres of macroalgae in Great Bay in 2007. DES then compared eelgrass coverage in 2007 to eelgrass coverage in 1996 (2421 acres) and by way of Figure 18 (see page 39 in DES 2009 Report) where they illustrated the spatial distribution of the 137 acres of macroalgal mat. Based on this analysis DES concluded that 5.7% of the area (137 acres ÷ 2421 acres) formerly occupied by eelgrass in Great Bay in 1996 was replaced where the median concentration of nitrogen was 0.42 mg N/L as determined by water quality monitoring surveys. Without providing quantitative criteria for determining what constituted significance, DES further concluded that the study by Pe'eri et al. (2008) showed that significant amounts of eelgrass are replaced by macroalgae when median total nitrogen concentration is 0.42 mg N/L.

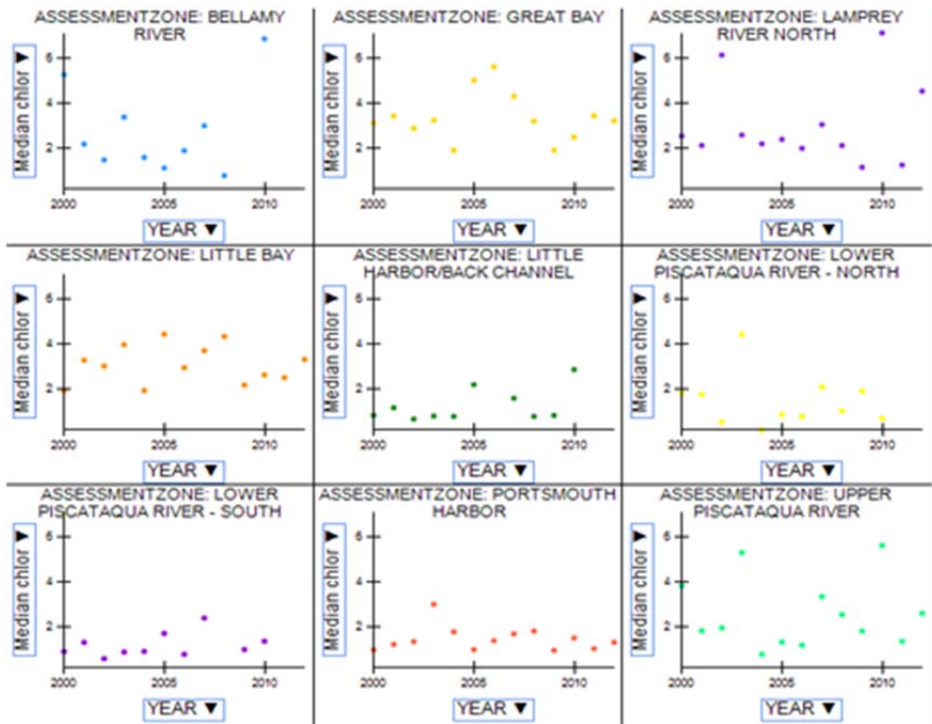
The data and arguments provided in the DES 2009 Report to support the weight of evidence for a relationship between nitrogen concentration, macroalgal abundance and eelgrass loss are neither compelling nor scientifically defensible. First of all, DES was inconsistent in their interpretation of what constitutes a significant eelgrass loss. In the original assessment of the status of eelgrass in the Great Bay estuary DES declared that a change would be considered significant if it was >20 %. The empirical basis for this value was based on the recorded variability of eelgrass cover in Great Bay as determined by monitoring surveys conducted between 1990 and 1999 (WD Doc R-WD-08-18, Methodology and Assessment Results related to Eelgrass and Nitrogen in the Great Bay Estuary for Compliance with Water Quality Standard for the New Hampshire 2008 Section 303(d) List). In their consideration of macroalgae DES did not explain why they concluded that a 5.7% change in eelgrass cover was significant when they had previously identified a 20% threshold for significance.

The second major problem with their evidence regarding the effect of macroalgae is that the data in the Pe'eri report are a study of a single year in one location in the Great Bay Estuary. On page 38 in their report DES correctly acknowledged it is not clear whether the same threshold would apply to other

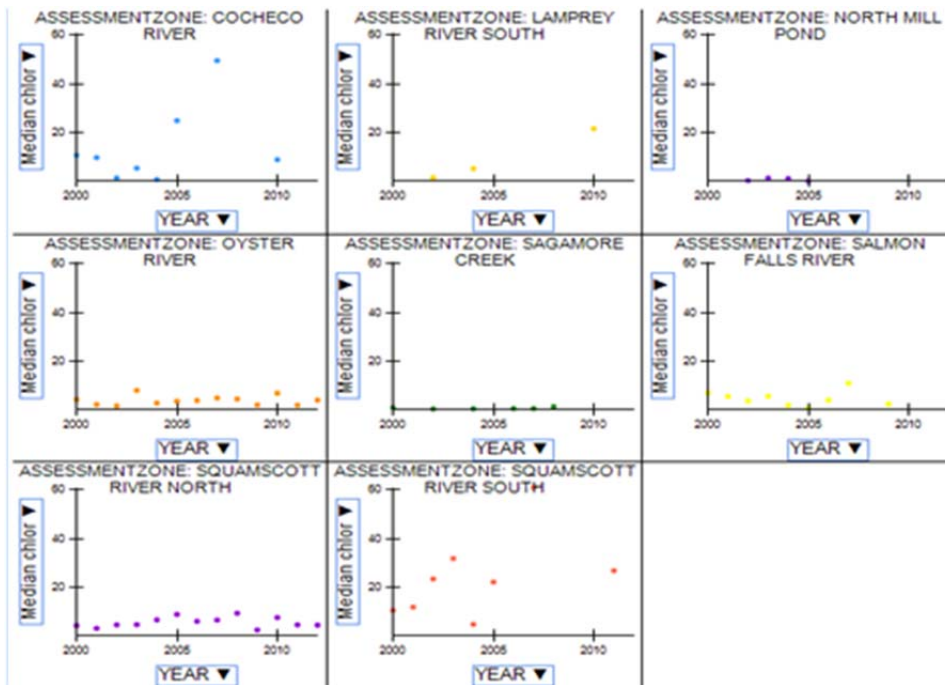
sections of the estuary where environmental conditions (e.g., substrate type, sediment stability, water depth, wave energy) may affect the growth and abundance of macroalgae and the interactions between macroalgae and eelgrass. Furthermore, a single year ($n=1$) cannot be considered representative of a highly dynamic shallow water macrophyte system consisting of eelgrass and macroalgae. Although DES has the capability to measure variation in eelgrass cover based on annual surveys (e.g., see Table 1), there are no data for variation in macroalgal abundance, so DES cannot determine if 2007 was representative of some average or median value for macroalgae in a longer time series. If 137 acres of macroalgae were having a permanent negative effect on eelgrass abundance in 2007, how do you explain the fact that eelgrass cover increased in Great Bay during 2008, 2009, and 2010 (Table 1)? In 2010 there were 477 more acres of eelgrass in Great Bay than present in 2007. This additional eelgrass cover is 3.5 times more eelgrass cover than was allegedly displaced by macroalgae according to the 2007 study. Inconsistency in the definition of what constitutes significance, the data for variation in eelgrass cover, and the extremely limited data for macroalgal cover and abundance renders any conclusions regarding nitrogen thresholds based on macroalgae effects unsupported.

RECKHOW RESPONSE

First of all, the NH data for chlorophyll do not present a time trend at the individual sampling sites; see the figures below. So, there is no chlorophyll time trend in the site-specific data, but there are spatial variations in chlorophyll among the sites. For the site-dates pooled data set, I plotted eelgrass coverage versus TN concentration for ordered categories of chlorophyll a concentration; this is presented in the next figure below. Note that there is no clear pattern between eelgrass and TN as we scan the plots from low to high chlorophyll. Thus, while I might expect that TN reductions will reduce chlorophyll levels, increase water clarity, and provide more light penetration for eelgrass growth, this is not supported in the cross-site data analysis.



A)



B)

Figure 5. Time Trends for Median Chlorophyll

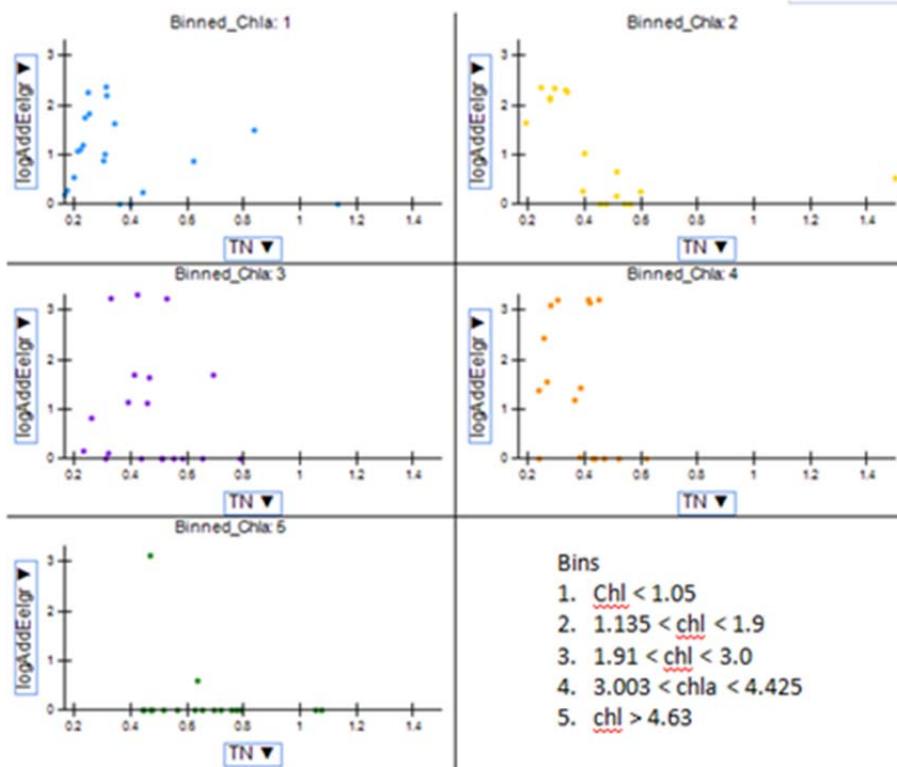


Figure 6. Eelgrass versus TN for categories on Chlorophyll

d) Does the available information indicate that algal growth is the reason for low DO conditions in the tidal rivers and that nitrogen reduction will significantly improve DO in the tidal rivers that flow into the Great Bay estuary?

BIERMAN RESPONSE

As stated above in my response to Question 1c, no data were presented for algal growth (primary productivity) in the DES 2009 Report. Phytoplankton are characterized exclusively by chlorophyll-a concentration.

With the exception of the nitrification process, nitrogen concentrations are not directly linked to DO, but are only indirectly linked through primary production and the subsequent sequence of physiological processes that utilize the produced organic matter. These include respiration, oxidation of DOC exudates, oxidation of POC, and sediment oxygen demand (SOD). Another necessary and confounding factor, with regard to lower DO, is physical stratification/vertical stability of the water column.

For the above reasons, development of scientifically credible statistical relationships between nutrient concentrations as a causal variable and DO as a response variable is difficult under any circumstances. In fact, even EPA itself was unwilling to demonstrate such a relationship in its own guidance. A notable omission, not generally recognized, is that the EPA Technical Guidance Document for Stressor-Response Relationships (EPA 2010b) does not contain a single example for dissolved oxygen as a response variable.

My opinion is that the results in Figures 28-29 of the DES 2009 Report for statistical relationships between DO and nitrogen concentrations, and the conclusions drawn from these results, are weak and unreliable because univariate linear regression approaches do not adequately represent the underlying direct/indirect cause-effect mechanisms. Conditions in Great Bay are driven by a set of physical, chemical and biological dynamics for which process-based mass balance models would be more appropriate tools for assessing water quality and resulting eutrophication. See my response to Question 4a for a more complete discussion.

DIAZ RESPONSE

Historically, there is no evidence of oxygen problems in the Great Bay region. In the mid 1980s when the first national assessment coastal waters was conducted there was no evidence of low dissolved oxygen (DO) conditions in Great Bay (Whitledge 1985) and the 2000-2001 National Coastal Assessment did not find indications of low DO in the New Hampshire Estuaries (EPA 2008). During the 2003-2006 National Coastal Assessment poor water quality was found in Great Bay (EPA 2012):

“...Pockets of poor water quality are apparent at stations in Great Bay, NH; Narragansett Bay, RI; Long Island Sound; New York/New Jersey (NY/NJ) Harbor; the Delaware Estuary; and the western tributaries of Chesapeake Bay. These hot spots largely reflect patterns of population density (see Figure 3-4) and industrial and agricultural activity in the Northeast.”

The direction taken in the DES 2009 Report relative to low DO appears logical given long-term trends in Great Bay, and what is known about other systems that have develop low DO and hypoxia from excess nutrient driven eutrophication.

Setting the threshold for low DO at 5 mg O₂/l at all times and 75% air saturation on a daily basis is a high bar that would be protective of all living resources within Great Bay. For coastal systems, 2 to 2.8 mg O₂/l is the point at which negative impacts on benthos are obvious (Diaz and Rosenberg 1995). The DES values for oxygen also mix concentration and partial pressure units. Depending on salinity and temperature, 75% saturation would range several mg O₂/l, for example for seawater from 10 to 30°C 75% saturation would be about 7 to 5 mg O₂/l (Figure 2).

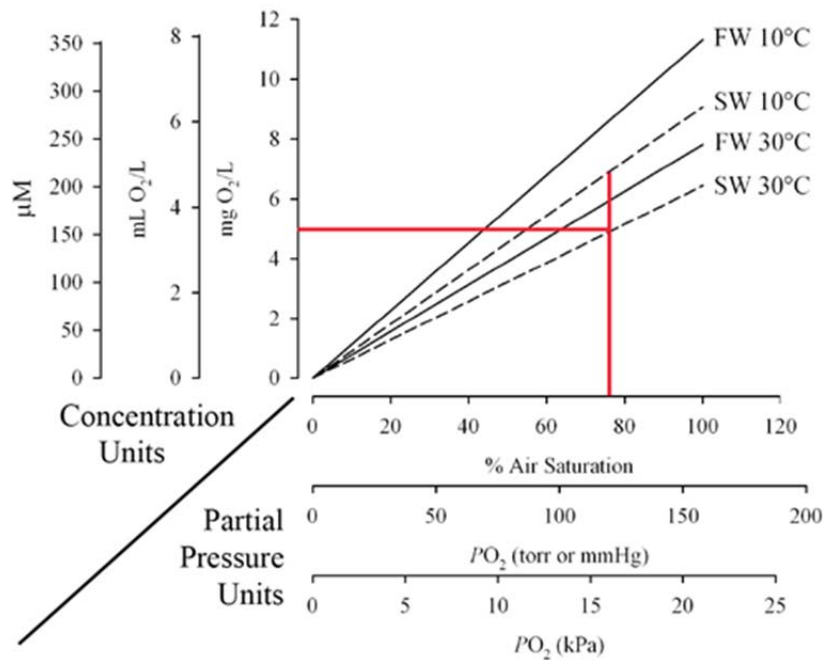


Figure 7. Nomogram for dissolved oxygen in fresh water (FW) and sea water (SW) at 10 and 30°C

Modified from Diaz and Breitburg 2009. Concentration units are on Y-axis, and partial pressure units are on X-axis. Red lines are the DES defined points for low DO

The combination of physical, chemical, and biological processes that lead to hypoxia differs in magnitude and importance by water body. For hypoxia to develop the amount of DO in the water column needs to be decreased by the process of respiration at a faster rate than resupply through atmosphere exchange, photosynthetic production of oxygen, or advection. Two factors must then be present for the development of hypoxia:

- Density stratification of the water column that isolates the bottom water from exchange with oxygen-rich surface water. Stratification is most commonly driven by salinity or temperature.
- Decomposition of organic matter in the isolated bottom water that consumes dissolved oxygen. In eutrophic systems, organic matter is supplied in excess of what the system is normally able to process.

Stratification of the water column is a physical characteristic of a system and must be present for low DO in the bottom water to develop. The role played by nutrients would be through enhancing the production of organic matter. Assuming the strength of stratification is beyond management, the goal of management would be to keep organic matter below the levels that would lead to low DO.

It is well known that excess algal (phytoplankton and macroalgae) growth can lead to hypoxia, but the question for algal growth and DO within the Great Bay system would be: Are the areas affected by low DO the same areas where the algal biomass production ends up? The most detailed information on macroalgal distribution in the DES 2009 Report is for Great Bay, but most of the low DO problems seem to be in the tributaries where chlorophyll-a, a measure of phytoplankton standing stock, tends to be higher (See DES 2009 Report figures 14, 18, and 25). If autochthonous primary production is the main source of organic matter to the Great Bay system and reductions in nitrogen loads lead to lower primary production then DO may improve. If allochthonous organic matter from terrestrial sources or municipal/industrial discharges is the main source, then the benefits of lowering nitrogen may be difficult to detect.

To assess if nitrogen reductions will improve DO conditions, data on the origin, quantity, and quality of organic matter in the various assessment regions of Great Bay are needed. In addition, the well-known relationship between sediment grain-size and organic matter has to be controlled (Hyland et al. 2005). Assessment region differences in depth, salinity, and sediment hydrodynamics also likely contribute to the linear regression relationship in many of the DES 2009 Report figures and need to be controlled. In particular, relating DO to nitrogen concentration as in figures 28 and 29 of the DES 2009 Report without accounting for the co-varying influence of these factors is too simple.

KENWORTHY RESPONSE

I defer to my colleague on the review panel, Dr. Diaz, who is an expert on the topic of dissolved oxygen in estuarine waters to provide a comprehensive response to this question. I do, however, reiterate what I indicated above with respect to macroalgal growth. Even though it is well documented that excessive growth of macroalgae could be responsible for low DO in a shallow estuarine system, the DES 2009 Report provides insufficient information on the distribution and abundance of macroalgae to link macroalgae to low DO and any implications for nitrogen reduction and eelgrass protection.

RECKHOW RESPONSE

Scientific understanding would support a positive answer to this question. I developed a causal Bayesian probability network to assess if the pooled sites-dates data support this statement. To understand the interpretation of a Bayes network, consider the figure below. This model describes linkages between variables, which are represented by the lines in the figure that connects the boxes. The model indicates that the node, or variable, “Chla” is conditionally dependent on the variables TN, Salinity, and SS. Similarly, the node “DO” is conditionally dependent on the node “salinity” alone.

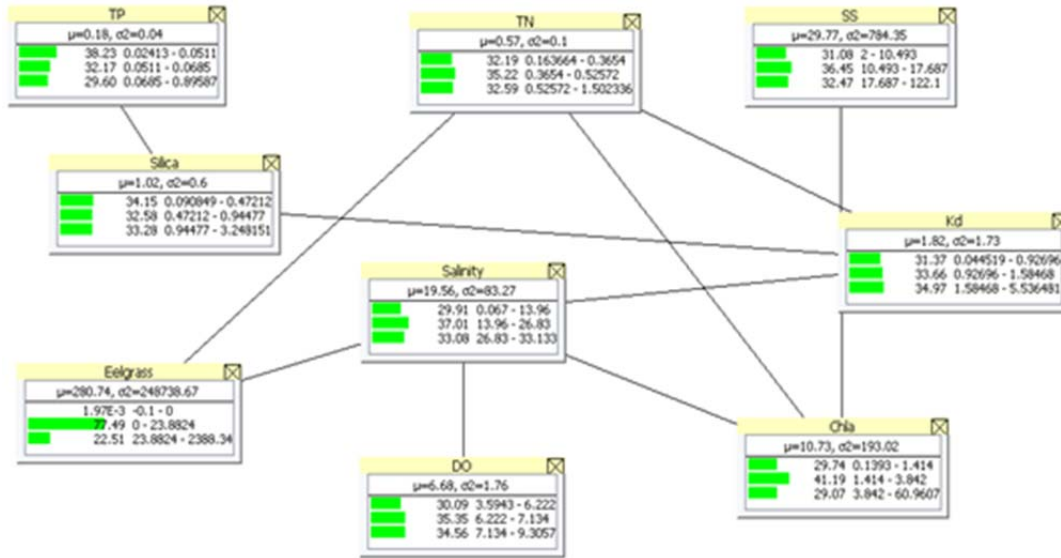


Figure 8. Baseline relationship among variables

The next figure shows how low DO and high TN affects eelgrass; low DO and high TN are represented by the red bars. The effect is relatively small as indicated by the change in the eelgrass bars between the two figures. This conclusion is demonstrated visually by how much the eelgrass green bars from Figure 8 are changed in Figure 9 for each category. The change in the length of the green bars even under the worst conditions for TN and DO is in the “expected” direction (toward lower eelgrass, as shown by the middle green bar in each figure), but it is small due to the limited available data and “noise” in the relationships.

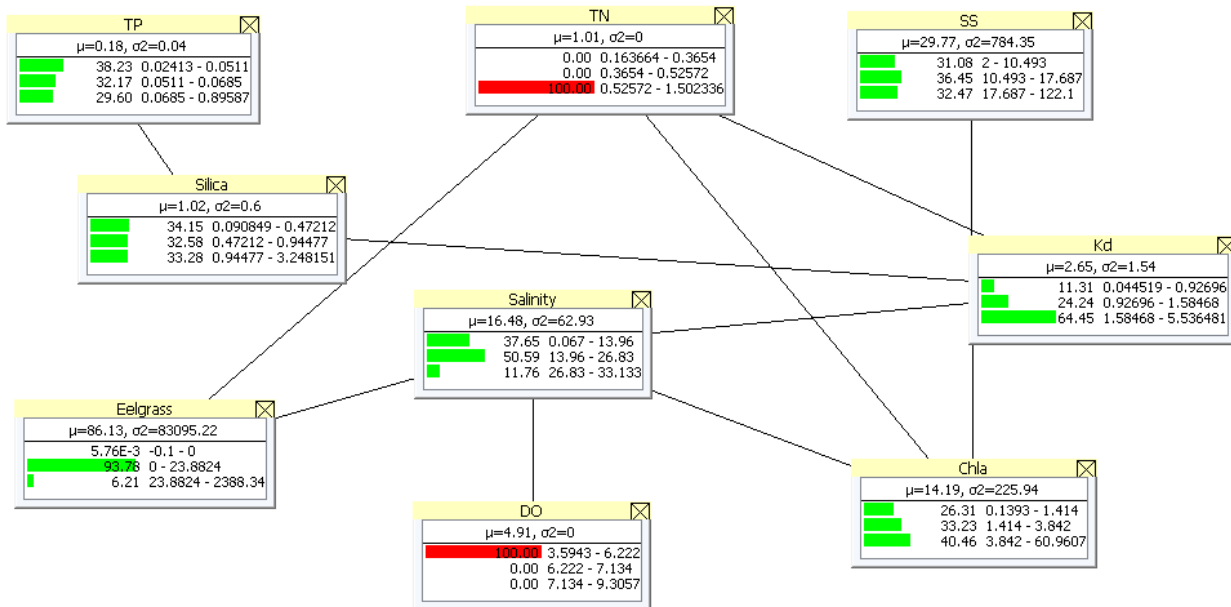


Figure 9. Effects of eelgrass from low DO and TN concentrations

e) Were the statistical methods used to derive the numeric thresholds based on acceptable scientific methods? Are the results of these analyses reliable for predicting responses to nitrogen in this system (including DO, transparency, eelgrass, macroalgae, phytoplankton, etc.)?

BIERMAN RESPONSE

The statistical methods used to derive the numeric thresholds were not based on acceptable scientific methods and the results of these analyses are not reliable for predicting the complexity of responses to changes in nitrogen concentration in the system, including DO, transparency, eelgrass, macroalgae and phytoplankton.

EPA (2010b), published 17 months after the DES 2009 Report, provides guidance for use of stressor-response relationships to derive numeric nutrient criteria. Key points in this guidance are the following:

- Statistical analyses can be applied to different types of waterbodies (including estuarine and marine systems) if sufficient data are available on causal variables, response variables and confounding factors
- A source of uncertainty in the accuracy of estimates of stressor-response relationships is the potential effect of environmental factors that co-vary with nutrient concentrations. These are called confounding factors and are environmental variables that also can influence the selected response variable. When the effects of a possible confounding variable are not controlled, the relationship estimated between the nutrient variable and the response variable may partially reflect the un-modeled effect of the confounding variable.
- The first step in evaluating the accuracy of a stressor-response relationship is to revisit the list of all possible confounding factors.

The DES 2009 Report did not adequately consider confounding factors in its SLR analyses. Some of these confounding factors include bathymetry, vertical stratification, currents, waves, tides, salinity, hydraulic flushing, phosphorus concentrations, total suspended solids (TSS), sediment GSD, sediment TOC and sediment sulfide concentrations. A detailed example is given below for salinity, but the same reasoning could be applied to these other confounding factors as well.

In the DES 2009 Report, the following SLR results are presented pertaining to benthic index of biological integrity (B-IBI):

- Figure 19: median B-IBI versus median TN, negative trend (R squared = 0.632)
- Figure 20: median B-IBI versus median salinity, positive trend (R squared = 0.638)
- Figure 21: median salinity versus median TN, negative trend (R squared = 0.680)

Page 40 of the DES 2009 Report states the following:

“While the B-IBI was well correlated with nitrogen concentrations (Figure 19), the best explanatory variable for B-IBI was salinity (Figure 20). Diversity and abundance of benthic infauna species are strongly affected by salinity. The B-IBI algorithm developed by EPA does

not correct for the effect of salinity on benthic community composition and is most accurate for higher salinity areas as discussed in Hale and Heltshe (2008). Therefore, the relationship between B-IBI and nitrogen concentration is probably just an apparent correlation caused by the inverse relationship of nitrogen and salinity in the estuary (Figure 21)."

This logic makes sense, but it is not clear why DES did not also apply it to their analyses of the other response variables in the report (chlorophyll, DO, turbidity, Kd). Figures 20 and 21 are the only SLR results in the report that explicitly include salinity. If the relationship between I-IBI and nitrogen concentration is, in fact, just an apparent correlation caused by the inverse relationship of nitrogen and salinity in the estuary (Figure 21), then why could this not also be the case for chlorophyll, DO, turbidity or Kd?

At my request, DES provided supplementary SLR results with nitrogen concentration replaced by salinity for Figures 15, 23, 28, 34 and 38 in the DES 2009 Report. In general, the strength of the correlations with salinity is comparable to those with nitrogen concentration. Given the strength of the correlation between salinity and nitrogen concentration (Figure 21; R squared = 0.680), this makes sense.

Several (some very literal) interpretations of the above results are possible:

1. The water quality/ecological health of Great Bay Estuary could be improved by increasing salinity, not by decreasing nitrogen.
2. Water quality/ecological health could be improved by decreasing nitrogen, not by increasing salinity.
3. Statistically, increasing salinity and decreasing nitrogen are the same remedies because they are strongly correlated.
4. In reality, increasing salinity and decreasing nitrogen are different remedies because the former would require alteration of freshwater inputs and/or tidal hydrodynamics, and the latter would require reductions in nitrogen loads from the watershed.
5. Salinity itself is not a causative factor, but an indicator of freshwater inputs, tidal hydrodynamics and hydraulic flushing rates, confounding factors that have not been explicitly considered in the analyses.

The importance of hydraulic flushing rates is also suggested by two other lines of evidence. First, the DES 2009 Report relied upon the conceptual model by Bricker et al. (2007). Although this model is presented in the form of a high-level pictorial and not a detailed schematic with all of the relevant stressor, response and confounding variables, it does contain explicit mention of hydraulic flushing as an influencing factor in Figure 4-1, a conceptual diagram of the North Atlantic. Second, on Page 17 of the DES 2009 Report it is stated that river inflow to the estuary is only 2% of the tidal prism exchange, and on Page 28 it is stated that most of the estuary (88% by volume) has salinity greater than 20 ppt. That is, more than half of the water in the estuary is ocean water. These statements not only point to the importance of considering how flushing rates differ among the 22 assessment zones, they beg the question of the load-response relationship for the system. For example, if nitrogen loads from the watershed are reduced, how much reduction in water column nitrogen concentration can be expected in Great Bay Estuary, and what would be the differences in responses among the 22 assessment zones?

My opinion is that DES should have first adopted a detailed conceptual model that explicitly represented all of the relevant direct/indirect causal linkages among stressor variables, response variables and confounding variables. Then DES could have used this conceptual model to design their statistical analyses to include causal linkages that make biological sense, not just statistical sense, and to explicitly account for co-varying/confounding factors. Instead, in most cases, they by-passed these critical steps and went directly to SLR analyses that presumed cause-effect relationships driven by nitrogen concentrations.

DIAZ RESPONSE

The statistical approach taken in the DES 2009 Report is understandable but fails to account for the complexity of interactions between response and explanatory variables. While EPA guidance on stressor-response relationships for deriving numeric criteria was not published till 2010, any further work on defining numeric nitrogen concentration criteria needs to conform to EPA guidance and best available statistical modeling.

The DES 2009 Report did not adequately consider confounding factors in its simple linear regression approach, which makes the interpretation and predictive ability of these regressions weak. For example, from figure 19 in the DES 2009 Report there appears to be an inverse relationship between median B-IBI and median TN, but salinity and sediment grain-size, two of the primary confounding factors that affect indices like the B-IBI were not controlled for. The DES 2009 Report does acknowledge that salinity is an important factor in assessing B-IBI and then points out the “...apparent correlation caused by the inverse relationship of nitrogen and salinity...”, but does not go further. Defining the relationship between B-IBI and any single water quality variable is difficult. In Chesapeake Bay it took a comprehensive multivariate approach to examine the relationship between a B-IBI and low DO (Dauer et al. 1992, Christman and Dauer 2003).

KENWORTHY RESPONSE

The statistical approach taken in the DES 2009 Report to derive the numeric thresholds for nitrogen were largely based on using simple linear regression to describe trends in total nitrogen concentrations and symptoms of eutrophication (e.g.; chlorophyll-a, K_d) and ultimately either the loss of eelgrass or the prevention of eelgrass recovery. The severe deficiency in this approach stems from the fact that it oversimplifies a stressor response in a highly complex estuarine ecosystem which may have important interactions between unaccounted for, and potentially confounding variables, such as temperature, salinity, currents, wave energy, bathymetry, water residence time, grazing, bioturbation, disease, and plant reproduction. From simple linear regressions you may be able to draw inferences about spatial or temporal trends in the relationships between variables (e.g., total N concentration and chlorophyll-a or K_d), but you cannot make strong inferences about cause and effect (stressor response) without empirical data to support the linkages. This is problematic for DES because there are no published empirical studies demonstrating the effects of total N on eelgrass which could be used to support their inference regarding a specific nitrogen concentration. There are numerous studies which have examined various forms of inorganic nitrogen (e.g., ammonium and nitrate) (e.g., Short and Burdick 1995, or see Burkholder et al. 2007 for a review) and the response of chlorophylls-a, epiphytes, light and eelgrass growth and biomass

that could be used to support relationships suggested by linear regressions. But DES does not develop a strong case for total N using these empirical studies. In fact, DES relies almost exclusively on citing total N values derived in Massachusetts embayments by the Massachusetts Estuaries Project (MEP) to support their determination of total nitrogen criteria. The value proposed by DES is similar to the range of values derived by the MEP. But the MEP used a reference condition approach supplemented and improved by linking mechanistic hydrological and watershed loading models to determine numeric total nitrogen criteria. The statistical approach taken by DES is much simpler than the MEP approach and it is difficult to support the proposed criteria because, as indicated earlier in my response to question #1, eelgrass is still declining in locations (reference conditions) with the lowest concentration of total nitrogen and the most transparent water. This would suggest that there are confounding factors affecting the response of eelgrass to the primary symptoms. The simple linear regression approach that identifies the upstream-downstream trend in total nitrogen and symptoms of eutrophication suggests this alternative conclusion, but more rigorous evaluation of the potentially confounding factors and use of more sophisticated statistical analytical and modelling tools are needed to develop strong inferences for setting nitrogen criteria.

RECKHOW RESPONSE

The statistical methods applied in the report are almost exclusively focused on univariate and bivariate relationships, yet the report writers recognize the multivariate nature of the relationships affecting eutrophication. Given that, and given my analyses on the multivariate patterns in the data, I do not think that the results in the report are acceptable or reliable for setting nutrient criteria.

QUESTION 2. THE DES 2009 REPORT USES A “WEIGHT OF EVIDENCE”. . . approach to identify a range of possible values for a TN threshold between 0.20 and 0.38 mg/L to protect eelgrass resources. TN thresholds of 0.25 to 0.30 mg/l were selected for areas with eelgrass, based on the regression of transparency to TN and depending on the restoration depth. THE DES 2009 REPORT selected 0.45 mg/L to maintain instantaneous do concentrations greater than 5 mg/L.

BIERMAN RESPONSE

No response to preface.

DIAZ RESPONSE

Weight of evidence is a reasonable approach to setting numerical nitrogen concentration criteria. This approach when combined with best professional judgment can be a powerful tool for drawing conclusions in many areas of water quality management. EPA (2011) issued guidelines on using weight of evidence in screening for endocrine disruptors. Much of the description of weight of evidence is applicable to numeric nitrogen criteria. EPA (2011) describes weight of evidence (WoE) as:

“Generally, WoE is defined as the process for characterizing the extent to which the available data support a hypothesis that an agent causes a particular effect (USEPA 1999; 2002a; 2005). This process involves a number of steps starting with assembling the relevant data, evaluating that data for quality and relevance followed by an integration of the different lines of evidence to support conclusions concerning a property of the substance. WoE is not a simple tallying of the number of positive and negative studies (US EPA 2002a). Rather it relies on professional judgment. Thus, transparency is important to any WoE analysis. A WoE assessment explains the kinds of data available, how they were selected and evaluated, and how the different lines of evidence fit together in drawing conclusions. The significant issues, strengths, and limitations of the data and the uncertainties that deserve serious consideration are presented, and the major points of interpretation highlighted.”

Part of the WoE approach is a set of general assessment factors to apply to the information used (EPA 2011):

- Soundness - Scientific and technical procedures, measures, methods or models employed to generate the information are reasonable for, and consistent with, the intended purpose.
- Applicability and Utility - The information is relevant for the Agency’s intended use.
- Clarity and Completeness - The degree of clarity and completeness with which the data, assumptions, methods, quality assurance, sponsoring organizations and analyses employed to generate the information are documented.
- Uncertainty and Variability - The uncertainty and variability (quantitative and qualitative) in the information or the procedures, measures, methods or models are evaluated and characterized.

- Evaluation and Review - The information or the procedures, measures, methods or models are independently verified, validated, and peer reviewed.

Overall, the DES 2009 Report does a good job with Soundness, and Applicability and Utility. Clarity and Completeness could be improved by applying more appropriate models that capture the complexity of interactions between nitrogen and assessment parameters. More could also be done with Uncertainty and Variability. Few of the data figures in the DES 2009 Report have variability estimators. When using median as the measure of central tendency, the range for the central 50% of data point should be included. Finally, the DES 2009 Report is under rigorous evaluation and comprehensive review.

KENWORTHY RESPONSE

No response to preface.

RECKHOW RESPONSE

No response to preface.

Specifically respond to the following:

a) Is “weight of evidence” a reasonable approach to selecting final thresholds for areas with eelgrass impairments and low do?

BIERMAN RESPONSE

The U.S. EPA Science Advisory Board (EPA 2010a), in its review of the August 17, 2009 draft technical guidance (the “Guidance”) on Empirical Approaches for Nutrient Criteria Derivation (EPA 2009), stated the following on “weight of evidence” approaches:

“When properly developed, statistical associations can be useful in supporting cause and effect arguments as part of a weight-of-evidence approach (further discussed in Section 3.3, recommendation #7 of this advisory report) to criteria development. Therefore, the final Guidance should provide more information on the supporting analyses needed to improve the basis for conclusions that specific stressor-response associations can predict nutrient responses with an acceptable degree of uncertainty. Such predictive relationships can then be applied, with mechanistic or other approaches, in a tiered weight-of-evidence assessment using individual lines of evidence in combination to develop nutrient criteria.”

“The Guidance should contain a quantitatively based weight-of-evidence framework using multiple methods and then combining them into figures and tables for visualization. Multiple statistical methods on one data set do not equate to a reasonable weight-of-evidence that significantly reduces uncertainty. Rather, the weight-of-evidence should involve different assessment methods (e.g., different data sets, different biological endpoints, measures of

habitat, etc.). This premise has been embraced by other EPA programs and the scientific community (Adams, 2003; Burton et al. 2002; Chapman, 2007; Chapman et al., 2002; Collier, 2003; Cormier et al., 2010; Fox, 1991; Linder et al., 2010; Linkov et al., 2009; Suter et al., 2002; Suter et al., 2010; U.S. EPA, 2000c; Weed, 2005; Wickwire and Menzie, 2010).”

“The Guidance can be used to develop numeric nutrient criteria in a tiered, weight-of-evidence assessment using appropriately modified EPA approved procedures together with other approaches that address causation. Large uncertainties in the stressor-response relationship and the fact that causation is neither directly addressed nor documented indicate that the stressor-response approach using empirical data cannot be used in isolation to develop technically defensible water quality criteria that will “protect against environmental degradation by nutrients.” The Guidance can, however, be used in a tiered, weight-of-evidence assessment (using appropriately modified U.S. EPA-approved procedures, e.g., EPA’s Causal Analysis/Diagnosis Decision Information System [CADDIS]), (U.S. EPA, 2009b).”

I am in agreement with these review comments by the Science Advisory Board, and my opinion is that “weight of evidence,” so defined, is a reasonable approach to selecting final thresholds for areas with eelgrass impairments and low DO.

EPA (2010b) (published 17 months after the DES 2009 Report) recommends three types of scientifically defensible approaches for developing numeric nutrient criteria:

- Reference condition approaches
- Stressor-response analysis
- Mechanistic modeling.

In this context, stressor-response analysis refers to empirical statistical analysis and mechanistic modeling refers to process-based mass balance water quality modeling.

The DES 2009 Report focused primarily on stressor-response analysis and secondarily on a reference condition approach. It did not use process-based mass balance modeling.

Development of numeric nutrient criteria involves establishing quantitative linkages between nutrient concentrations and direct response variables such as chlorophyll-a concentrations, and indirect response variables such as DO and eelgrass. These are complex relationships and require explicit consideration of many co-varying/confounding variables. My opinion is that a scientifically defensible “weight of evidence” approach to development of numeric nutrient criteria should include lines of evidence based on all three of the above approaches recommended in EPA (2010b).

Page 66 of the DES 2009 Report summarizes the “weight of evidence” used to determine a nitrogen threshold for protection of eelgrass. Although data from other estuarine/marine systems were cited, this line of evidence was not sufficiently detailed to constitute a comprehensive reference condition approach. As stated above in my response to Question 1e, the statistical methods used in the DES 2009 Report for stressor-response relationships did not follow accepted scientific methods, and the results of these analyses are not reliable for predicting responses to changes in nitrogen concentrations in the system.

Although EPA has not developed technical guidance for use of process-based mass balance models to develop site-specific numeric nutrient criteria, such guidance is now available in a report by the Water Environment Research Foundation (Bierman et al. 2013).

DIAZ RESPONSE

Weight of evidence is a reasonable approach for evaluating causes and effects of low dissolved oxygen. As stated in the DES 2009 Report: “For aquatic life use support, DES investigated nutrient thresholds for the protection of the benthic invertebrate community, dissolved oxygen, and eelgrass.” It is typically assumed that low DO poses an immediate impairment threat primarily to benthic invertebrate communities, but low DO may also directly impair eelgrass physiology (Holmer and Bondgaard 2001). In addition, low DO can lead to cascading effects by releasing eelgrass epiphytes from invertebrate grazing pressures, further stressing eelgrass (Moksnes et al. 2008, also see Kenworthy response).

Relative to benthos, samples collected in the Great Bay system by the National Coastal Assessment were used to characterize benthic communities, total organic carbon content of the sediment, and sediment grain-size. From these data a benthic index of biologic integrity (B-IBI) developed by the Atlantic Ecology Division of EPA for the Gulf of Maine was calculated. This B-IBI is based on a multiple linear regression with three variables. The B-IBI increases with higher Shannon-Wiener H' diversity and mean of 5th percentile of total abundance frequency distribution of each species in relation to its ES50 value (Figure 3). The B-IBI decreases as the percent abundance of capitellid polychaetes increases:

$$\text{B-IBI} = 0.494 * \text{Shannon} + 0.670 * \text{Mean_ES50.05} - 0.034 * \text{PctCapitellidae}$$

Benthic community conditions were considered poor if the B-IBI was less than 4.

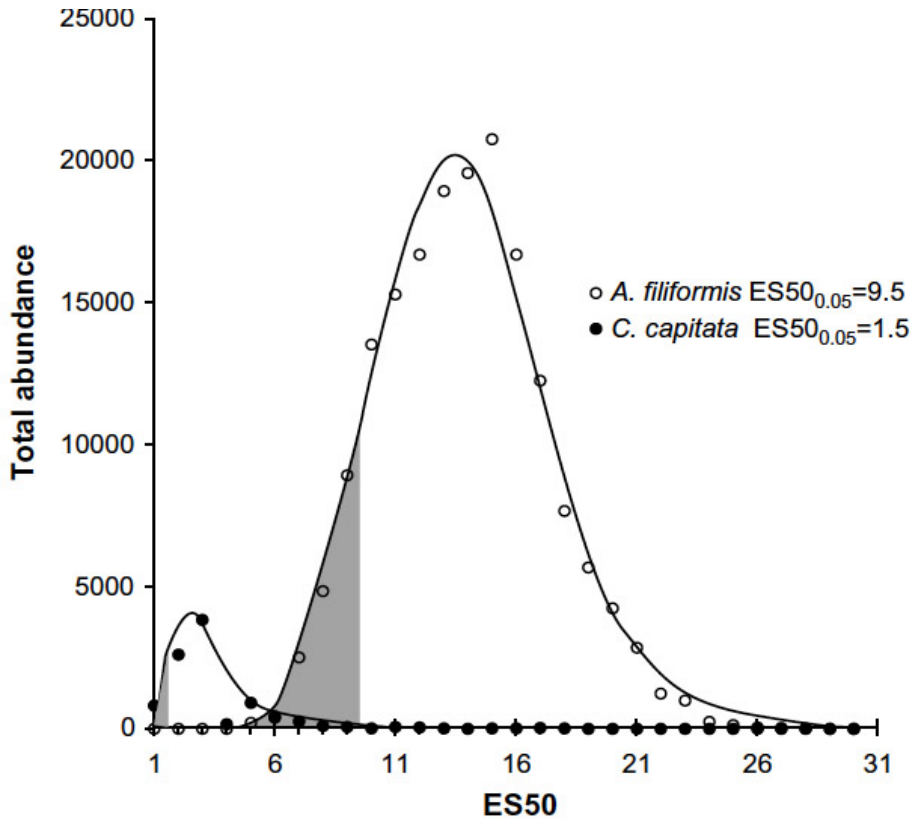


Figure 10. Examples of total abundance frequency distributions of the capitellid polychaete *Capitella capitata* and the brittle star *Amphiura filiformis* in relation to the expected number of species per 50 individuals (ES50) values for all samples in which they occur.

Shaded areas indicate the 5% abundance distribution in relation to the lowest ES50 values (ES50.05); for *C. capitata* 1.5 and for *A. filiformis* 9.5. Tolerant species like *Capitella capitata* are by definition predominantly found in disturbed environments and they would mainly occur at stations with low ES50. In contrast, sensitive species like *Amphiura filiformis* occur in areas with no or minor disturbance and would then be associated with high ES50 (Rosenberg et al. 2004).

These three benthic variables (B-IBI, Sediment TOC and grain-size), plus salinity, are a start for the weight of evidence approach to assess benthic conditions relative to nitrogen concentration and low DO. Additional variables that would be required included season and year. From the DES 2009 Report, it is not possible to determine the quantity of data that went into investigating nutrient thresholds for the protection of the benthic invertebrate community, dissolved oxygen, and eelgrass. As stated, benthic data from approximately 130 station visits in the Great Bay Estuary collected by the National Coastal Assessment from 2000 through 2005 were used. From examining available data for Great Bay Estuary over this time period, it is not possible to determine how the data were condensed into the 11 or 12 assessment region points in figures 19 to 23. Which of the stations listed by the National Coastal Assessment for the region in figure 4 were used in the DES 2009 Report? More detail on which assessment regions ended up with and without data is needed. Also are the sediment and water column samples synoptic and from the same location? Combining datasets collected for differing objectives is difficult and must be done with clear statement of strengths and limitations.

data used in the DES 2009 Report, between 2000 and 2012 the minimum DO concentration was 5 mg O₂/l or less a total of 20 times in eight of the 22 assessment zones. It therefore seems unlikely that low DO is a controlling factor for benthic community structure.

KENWORTHY RESPONSE

Yes, a “weight of evidence” approach is a reasonable means of selecting final thresholds for areas with eelgrass impairment as per EPA technical guidance (EPA 2009, EPA 2010a). The EPA documents provide specific guidance and recommendations on using multiple assessment methods with quantitative metrics, different data sets, and rigorous analytical statistical techniques. Unfortunately, in the case of the DES assessment in Great Bay, these guidance documents post-date the DES studies and the 2008 and 2009 DES reports. DES could not directly benefit from recommendations and guidance provided by these documents; however, it is my opinion that DES could improve their weight of evidence approach by implementing the recommendations made by EPA in these more recent guidance documents. This may require additional data collection, analyses of more recent water quality and eelgrass data, and the incorporation of mechanistic models into their assessment.

RECKHOW RESPONSE

In a general sense, “weight of evidence” is always a reasonable approach. In a specific sense, it depends on the evidence and on how the evidence is interpreted.

b) Does the “weight of evidence” (i.e., an assessment of available data and studies for this estuary) support the conclusion that excess nitrogen was the primary factor that caused (1) the decline of eelgrass populations or inability of eelgrass to repopulate specific areas, and (2) low DO in the tidal rivers?

BIERMAN RESPONSE

I defer to Drs. Diaz, Kenworthy and Reckhow.

DIAZ RESPONSE

The use of static indices like the B-IBI, in which the sensitivity to stressors does not account for possible shifts in sensitivity of species along natural environmental gradients, to assess any one individual stressor, such as DO is questionable. This is even more difficult given the limited range of DO conditions reported for Great Bay. The evidence presented that links B-IBI to total nitrogen concentration is not convincing. The DES 2009 Report properly interprets the complexity of the relationship between co-varying factors, B-IBI, and nitrogen, but it fails to follow through with a similar evaluation relative to B-IBI and DO. This leads the DES 2009 Report to set a total nitrogen concentration for keeping DO above the standards of 5 mg O₂/L at all times and daily average saturation at least 75% that is not supported by either a stressor-response or weight of evidence approach.

Relative to weight of evidence, the data presented are likely sound but are not properly applied to linking benthic conditions with low DO and subsequently to linking low DO with total nitrogen concentrations. Much of the problem is with the analysis approach being limited to simple linear regressions, which do not properly evaluate the influence of co-varying factors that confound conclusions regarding total nitrogen concentration as being the causal factor for DO and benthic conditions.

KENWORTHY RESPONSE

The DES 2009 Report explicitly states; *“The nitrogen threshold for the protection of eelgrass was derived using a weight of evidence approach which included the thresholds for macroalgae proliferation, regressions between total nitrogen and the light attenuation coefficient, offshore water background concentrations, reference concentrations in areas of the estuary which still support eelgrass, and the thresholds that have been set for other New England estuaries.”* Based on my responses to question #1 it is my opinion that the DES “weight of evidence” does not support the conclusion that excess nitrogen was the primary factor that caused the decline of eelgrass and the inability of eelgrass to repopulate specific areas.

RECKHOW RESPONSE

The PREP 2009 report provides additional years of data than available in the 2009 Numeric Nutrient Criteria (NNC) report. With more years reported, the PREP report shows more downward trend in

eelgrass coverage than does the NNC report. The two figures shown here based on the NNC report indicate the limited basis for eelgrass trends determination.

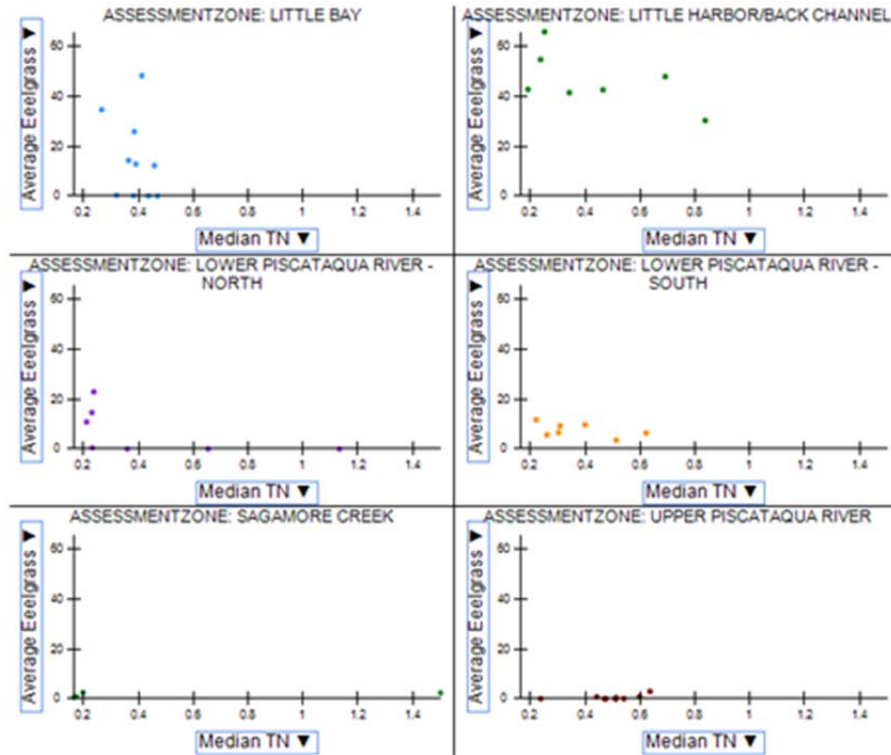


Figure 12. Eelgrass – TN relationship at monitoring stations

The NNC data for Little Harbor/Back Channel provide the best evidence for a downward trend in eelgrass coverage. However, the next two figures do not present a very strong case for eelgrass response to TN changes.

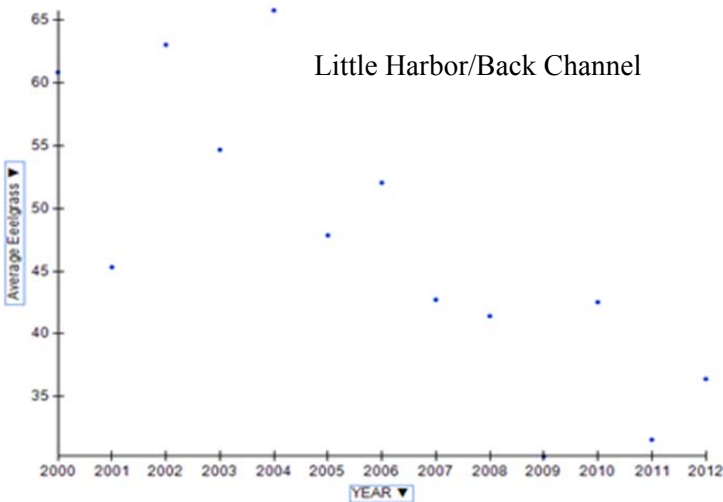
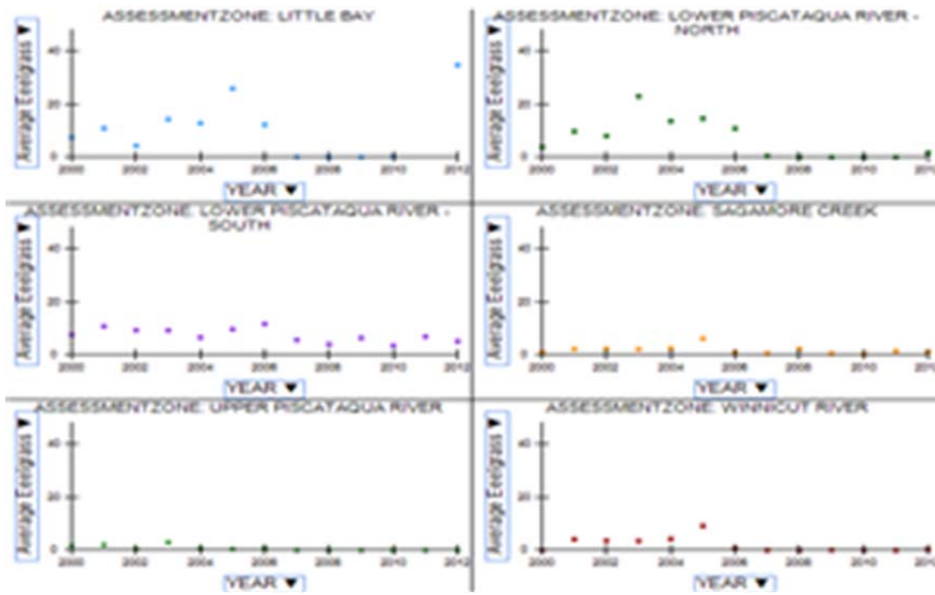
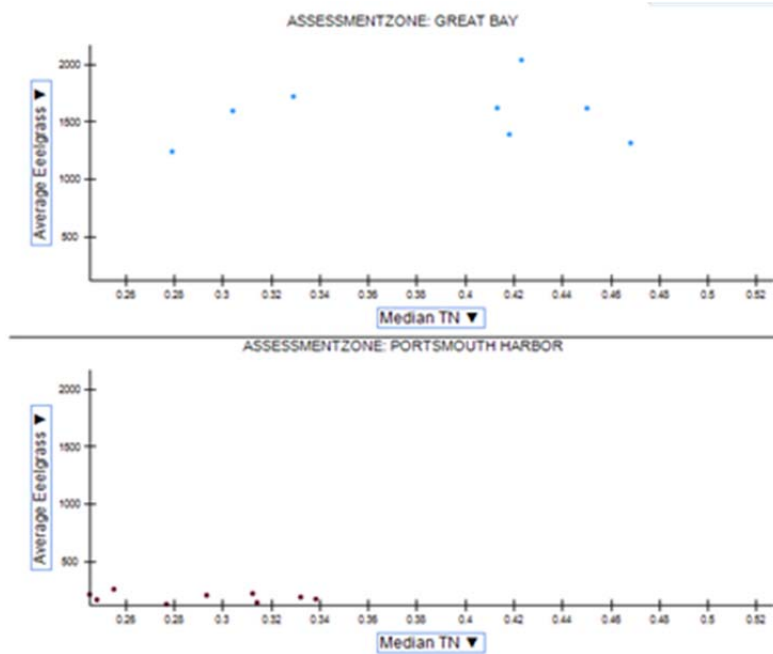


Figure 13. Downward trend in eelgrass coverage for Little Harbor/Back Channel

Scientific knowledge indicates a causal linkage between TN and DO, due to the growth and decomposition of algae. However, as I indicated in my response to Question 1d, my data analysis does not support this TN-DO linkage in the NH DES data.



A)



B)

Figure 14. Time trends for eelgrass at monitoring stations

c) Does the DES 2009 REPORT and/or subsequent data reasonably assess the potential reasons for eelgrass loss besides cultural eutrophication in the various areas?

BIERMAN RESPONSE

I defer to Dr. Kenworthy.

DIAZ RESPONSE

I defer to Dr. Kenworthy.

KENWORTHY RESPONSE

The answer to this question is no, as explained in my response to question #1. In particular, many of the potentially confounding factors known to affect eelgrass growth, reproduction, abundance, distribution and survival were not adequately ruled out as reasons for eelgrass declines. Furthermore, eelgrass cover data subsequent to the DES 2009 report (Table 1) indicates eelgrass is declining in locations (reference locations) where the nitrogen concentrations are similar to the proposed criteria; hence other factors must be operating to affect the changes in eelgrass cover.

RECKHOW RESPONSE

I defer to Dr. Kenworthy.

d) Are the selected TN criteria for eelgrass protection consistent with (1) data/studies available for this estuary and (2) TN levels found to be protective in other northeast estuarine systems?

BIERMAN RESPONSE

I defer to Drs. Kenworthy and Reckhow.

DIAZ RESPONSE

I defer to Drs. Kenworthy and Reckhow.

KENWORTHY RESPONSE

On page 3 the DES 2009 Report explicitly states that numeric nutrient criteria have been established for relatively few estuaries and they typically fall between 0.35 and 0.49 mg N/L, but they do not cite any specific reports or publications to document these values. DES refers to a total nitrogen criterion (0.49 mg N/L) adopted for Pensacola Bay in Florida, but this is a subtropical seagrass system with very different species and would not necessarily apply to eelgrass in the Great Bay estuary. As a precedent for eelgrass and total nitrogen criteria, DES explicitly identified the Massachusetts Estuaries Project (MEP) and the similarity between the range of values determined by MEP (0.30 – 0.38 mg N/L) and the proposed DES nitrogen criteria.

DES was correct in considering the MEP program as precedent because; 1) eelgrass is the primary species of interest, 2) eelgrass declines are well documented, 3) nitrogen is implicated as a stressor causing the declines, and 4) many of the bio-physical characteristics of the coastal ecosystems are similar. However, DES failed to acknowledge the relevance of some very important differences between the MEP program's approach and the DES approach. Also, important differences in some the physical characteristics of Great Bay and the embayments of Massachusetts were not acknowledged, implying that DES did not consider the relevance of the differences and how they could affect interpretation of water quality monitoring data. Furthermore, by making a simple comparison to the MEP program without a comprehensive evaluation of the status of that program, DES was irresponsible in making the comparison and implying that it supports total nitrogen criteria proposed for the Great Bay. None-the-less, consideration of the MEP program can inform DES in revising and improving their approach to setting nitrogen criteria.

The MEP program developed nitrogen criteria using two of the three approaches recommended in the most recent EPA guidance; 1) reference condition (DES refers to this as sentinel sites) and 2) mechanistic modelling. The MEP approach correctly recognized that because of the wide range of biophysical characteristics in coastal MA, as well as the different features of the watersheds, they could not set one criterion for all 89 embayments in MA. The MEP proposed assessing and modelling each embayment's hydrodynamics, watershed and nitrogen processes separately. MEP has completed assessments and modelling in a subset of the 89 embayments. Since it has been determined that the primary driver for eutrophication and eelgrass loss in many of the MA coastal embayments on Cape Cod, the Elizabeth Islands and Buzzards Bay is nitrogen enrichment from groundwater, the MEP linked watershed

embayment models address this process as well as other nitrogen transformations to model nitrogen criteria indicated by “healthy eelgrass” growing at reference stations. The approach has gone through scientific peer review and the initial stages of the implementation of nitrogen TMDLs for a small subset of the 89 individual embayments and achievement of the total nitrogen criteria has been initiated. The implementation of TMDLs and achievement of nitrogen criteria is at a very early stage in MA. It will take years, perhaps even decades, to finance and make the necessary infrastructural changes to modify the delivery of nitrogen to the groundwater. Even when that is accomplished the legacy of nitrogen already in the groundwater of many of the watersheds will take several years to decades to be depleted and eventually to detect improvements in the conditions of the embayments. By no means has the modeled and proposed nitrogen concentrations and expected eelgrass responses been tested and validated by MEP in any of the MA embayments.

It is my opinion that a simple comparison of total nitrogen values derived in the MEP cannot support the nitrogen concentration proposed by DES. To the best of my knowledge, I am unaware of any other northeast State that has total nitrogen criteria intended for the protection of eelgrass. None-the-less, DES could benefit from a more comprehensive evaluation of the MEP program and adopting at least some of the basic principles of the approach for the Great Bay assessment. For example, MEP recognized the distinct biophysical and chemical differences between watershed/embayment systems and assessed them separately.

The principle ‘no one suit fits all’ was applied appropriately in MA. This resulted in some embayments having different nitrogen criteria in MA, and recognition that no one concentration value will fit for all of the different systems. Although DES explicitly recognizes different segments of the Great Bay estuary, in order to discover nitrogen criteria the method DES used failed to consider potentially important differences that could affect nitrogen, symptoms of nitrogen loading, and the eelgrass response. For example, the lower salinity tributaries of Great Bay have distinctly different biophysical characteristics and much tighter coupling to the watersheds than further downstream which is more coupled to oceanic influences. There may in fact be some situations upstream in lower salinity where phosphorus is a controlling factor. It is also clear from the eelgrass cover data that some portions of the Great Bay should be considered largely a restoration problem (e.g., Winnicut, Squamscott, Lamprey and Oyster Rivers), while other locations would be considered primarily a maintenance and conservation problem (e.g., Great Bay, Portsmouth Harbor). It is likely that the eelgrass water quality requirements (light especially) and nitrogen criteria could be different in these locations. Restoration site selection criteria described by Short et al. (2002) suggest a number of factors that should be considered in the assessment of the different zones of Great Bay.

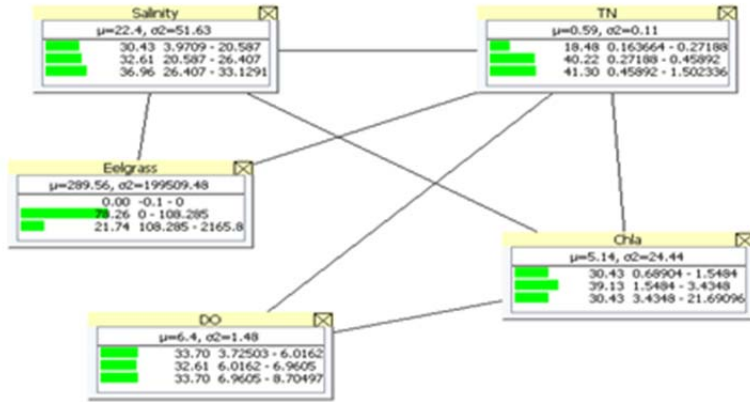
Considering the MEP approach identifies another very important deficit in the DES 2009 Report. With the exception of what was considered as the oceanic boundary conditions in the Gulf of Maine, DES did not take into account the inputs of nitrogen (loads and sources) and the potential variability of these inputs into the Great Bay estuary, better yet, different inputs in the various zones. Loading models would be very informative for the process of establishing achievable nitrogen criteria.

To the best of my knowledge, only one system wide level study of the relationship between total nitrogen and eelgrass status has been published in the scientific literature (Wazniak et al. 2007). This study was conducted in the coastal bays of Maryland and Virginia and examined the long-term record for trends in eelgrass abundance and total nitrogen concentrations, chlorophyll a, total phosphorus, and dissolved oxygen). This study is informative for DES because it demonstrates statistically that in locations where total nitrogen concentrations exceeded 0.65 mg/l eelgrass was declining. The proposed DES total nitrogen criteria in Great Bay (annual median of 0.25 – 0.30 mg total nitrogen) are about half the threshold concentration identified by Wazniak et al. (2007), so it appears that the DES criteria are more conservative and potentially more protective of eelgrass than identified for the Maryland coastal bays.

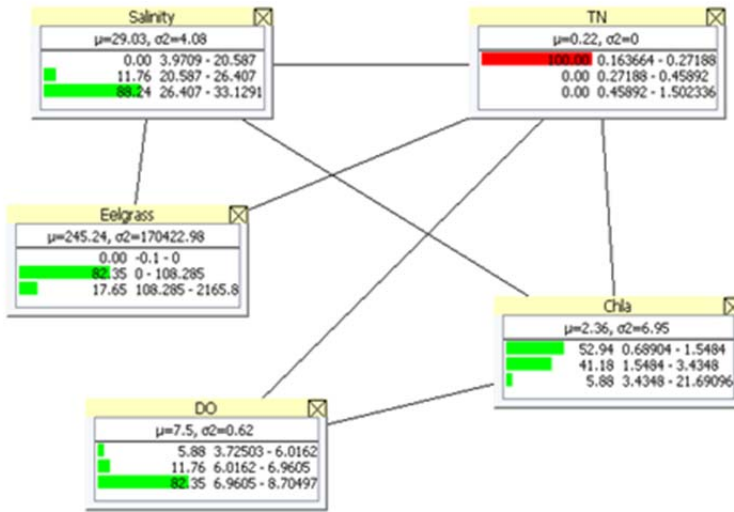
To help better identify the potential total nitrogen criteria for Great Bay, DES should also consider the results of a recent study conducted in collaboration with the MEP program in Massachusetts (Bensen et al. 2013). This study identified; 1) healthy and stable eelgrass beds as locations with long-term total nitrogen concentrations (2000-2010) of 0.45 mg/l and, 2) degrading or lost beds with concentrations of \approx 0.55 and 0.65 mg/l, respectively (see Figure 2 in Bensen et al. 2013). These results corroborate values reported by Wazniak et al. (2007) discussed above, indicating that concentrations on the order of about 0.6 mg/l total nitrogen correspond with degrading eelgrass beds. However, as indicated above in my responses to questions #1 and #2, even where lower total nitrogen values in Great Bay are lower than 0.6 mg/l and are at the proposed DES criteria concentrations, eelgrass is declining. Again, suggesting the likelihood that other factors are affecting eelgrass distribution, abundance and survival in Great Bay.

RECKHOW RESPONSE

Looking at only the pooled site/date NH DES data, I developed the Bayes network in the next figure. The cutoffs for the “bins” for the TN variable were selected based on the proposed TN criteria. To assess the impact of the proposed TN criteria, we should compare the changes in the three eelgrass bars from the “base” case in the first figure below to the eelgrass bars in the next two figures. In the second figure, I examined the impact on eelgrass of $TN < 0.27$ (indicated by the red bar); note that the change in eelgrass coverage from the base case is a relatively small improvement, as the middle category for eelgrass increases from 78% to 82%. In the third figure, I examined the impact on eelgrass of $TN > 0.45$ (indicated by the red bar); note that the change in eelgrass coverage from the base case is actually counterintuitive, as the middle category for eelgrass increases from 78% to 94%. Note however, that the changes in DO and chlorophyll **are** consistent with scientific understanding. Taken together, these findings underscore the weak link between TN and eelgrass in the NH DES data; this is consistent with other analyses I have presented.



A)



B)



C)

Figure 15. The predicted effect of the one of the proposed TN water quality criterion

e) Does the available information demonstrate that, for the protection of eelgrass habitat, the annual median total nitrogen concentration should be less than or equal to 0.25-0.30 mg N/L, depending on the eelgrass restoration depth? Will attaining these values achieve the desired restoration depth for transparency?

BIERMAN RESPONSE

I defer to Drs. Kenworthy and Reckhow.

DIAZ RESPONSE

I defer to Drs. Kenworthy and Reckhow.

KENWORTHY RESPONSE

First of all, there is compelling scientific evidence that eutrophication of estuaries and coastal embayments and loss of eelgrass can be caused by either the loading or delivery of high concentrations of different forms of inorganic, organic, and total nitrogen (e.g., Taylor et al. 1995, Short et al. 1995, Short and Burdick 1996, Kemp et al. 2004, Burkholder et al. 2007, Krause-Jensen et al. 2008, Vaudry et al. 2010, Latimer and Rego 2010, Benson et al. 2013). Several of these studies also make a direct link between nitrogen concentrations, nitrogen loading and water transparency. Likewise, eliminating point source wastewater discharges and reducing nitrogen loading reversed eelgrass losses in a shallow coastal embayment on Long Island Sound, Ct (Vaudry et al. 2010). Lending credence to the argument that nitrogen management can improve water quality conditions (e.g., water transparency) for the protection and restoration (Dennison et al. 1993, Krause Jensen et al. 2008, Vaudry et al. 2010). None of these studies actually specify any threshold concentrations of total nitrogen, and most either directly address concentrations of inorganic nitrogen (ammonium, nitrate/nitrite, phosphorous), or nitrogen loading.

DES was correct in considering measurements of water transparency, because it is a very important symptom of eutrophication and one of several factors controlling eelgrass distribution and abundance. As explicitly stated in the 2009 Report, DES quantified transparency as the light attenuation coefficient (K_d) derived from a number of data sources, presumably all using similar methods. To predict the presence or absence of eelgrass in different zones of the Great Bay estuary DES adopted a modelling approach suggested by Koch (2001) which incorporates the effects of tide range and assumed a fixed eelgrass light requirement (22%). The model was derived from a published empirical study of eelgrass depth distribution in Long Island Sound (Koch and Beer 1996) and DES used eelgrass light requirements (22%) adopted by the Chesapeake Bay Program. I am unaware of any studies which have rigorously tested or applied the Koch (2001) model for deriving nitrogen criteria; however, the model is based on sound physical principles, was derived in a relatively similar northeastern coastal environment (Long Island Sound) and should have practical value in the Great Bay estuary.

The assumption that seagrass light requirements for a species are constant, even in different environmental conditions, has come under scrutiny in several studies (Kenworthy and Fonseca 1996,

Duarte et al. 2007, Kenworthy et al. 2013). Light requirements may vary as a function of optical water quality (turbidity, transparency), sediment organic matter content (sulfide toxicity, oxygen demand) and water temperature. A recent study in coastal Massachusetts indicates that eelgrass minimum light requirements can range from as low as 9.6% in a pristine embayment to as high as 29.7% in a nitrogen impaired site (Kenworthy et al. 2013), suggesting it may be necessary for DES to evaluate the strength of the assumption of a fixed light requirement for eelgrass in Great Bay. DES should also consider what factors might affect their assumption (e.g., sediment organic matter, water turbidity, CDOM) and whether there is uncertainty in the assumed constant. DES should also explain why they depended on a value derived from the Chesapeake Bay Program, but neglected to specifically cite the results of a very relevant local empirical study by Short and Burdick (1995).

In a controlled mesocosm study at the University of New Hampshire Lab on Great Bay, Short and Burdick (1995) examined the effects of eutrophication and shading on eelgrass. This is an important study because, to a large extent, the results were not confounded by other factors that cannot be controlled in field surveys. This study identified a minimum eelgrass light requirement ranging between 11% and 21%. In the 21% light treatment, eelgrass densities were “steady” near the end of the experiment while at 11% they were still declining. The authors concluded that at the lowest level of light (11%) eelgrass could not be sustained. However, at values above 21% eelgrass growth and density increased. Based on this study, it would seem that 22% is a reasonable estimate of a “minimum” light requirement, but the plants grow and reproduce at higher light levels. The study is informative because it demonstrates that using a designated minimum value as a target may be a risky proposition, should some other factors stress the plants. In the experiment, at 21% the plants were surviving but were poised at a tipping point where they might be nudged to decline with less light or increase with more light, depending on what other stressors might affect them (e.g., temperature, nutrients, sediment organic matter).

A more recent mesocosm study conducted at the University of New Hampshire facility on Great Bay in collaboration with another study at the University of Rhode Island on Narragansett Bay also indicates that the light requirements of eelgrass may be higher than 22% (Short et al. 2012). In this study, plants grown at 50% of ambient light in Great Bay water exposed to high organic matter sediments (8%) and temperatures elevated 2^o and 4^o C above ambient Narragansett Bay water displayed significantly greater stress responses compared to plants grown at ambient light and temperature (e.g., depressed shoot growth, slower asexual reproduction rate). These empirical studies suggest that eelgrass light requirements are not constant and important interactions between other factors that affect eelgrass growth and reproduction should be considered in order to establish an accurate and protective light requirement for the plants.

It would also make sense that, in order to have a protective target value, it should be greater than the minimum. Most of the data supporting a 20-22% minimum value are derived from field studies at the deep edge of established eelgrass beds and the correspondence between the percentages of surface light reaching those edges. In many cases, these studies have been conducted in relatively healthy eelgrass beds where the plants are reproducing and clonal integration between plants is supporting growth at the deep edges. This is especially relevant because several of the tributaries in Great Bay have lost all or most of their eelgrass and it is generally understood that eelgrass light requirements in recovery conditions (e.g., a restoration or by natural seed recruitment) will be higher than at the edge of an established and healthy

meadow. DES acknowledged this possibility in the 2009 report, but it was not addressed directly and 22% was assumed for the entire system. Just as they did for setting the nitrogen concentration criteria values, DES should consider the uncertainty in eelgrass light requirements and the fact that a minimum may not be conservative enough to protect and restore the plants. DES should consider building in a “margin of safety” by assuming a higher value that would better ensure the growth, reproduction and expansion of eelgrass, and not just survival at a minimum threshold. Based on the studies cited above, it is also probable that eelgrass light requirements could vary in the different segments of the Great Bay estuary, especially since many areas are going to require restoration and not just maintenance. DES acknowledges these issues pages 56 and 57 of the 2009 Report, but they do not make any effort to address their implications.

DES was correct in using the K_d values to help determine a target depth for eelgrass bed maintenance and restoration in the different zones of the Great Bay estuary. This general approach has frequently been used by scientists and resource managers to establish goals for seagrass conservation (Orth et al. 2010 a, b). However, a more useful and quantitative approach would also take into account the; 1) actual distribution of eelgrass with respect to depth, and 2) the potential eelgrass distribution with respect to depth (see Wazniak et al. 2007) . By simply setting a target depth based on K_d and not having any information of the estuary’s bathymetry, it is impossible to determine what the implications of the target depth criteria will be with respect to the distribution and abundance of eelgrass. Without information on bathymetry I don’t know if and where those water depths occur in a zone, and how much of the zone would actually be suitable for eelgrass growth. The assessment approach should also include spatially articulated estuarine bathymetry information. Another assumption DES makes in their approach, but fails to address, is whether the target depth will support eelgrass. Are the substrate and environmental conditions at the proposed target depths throughout a zone suitable for eelgrass growth? This is an important question that should be acknowledged and addressed by DES before anyone can fully understand and predict the implications of the proposed criteria.

RECKHOW RESPONSE

See my response to part d above.

QUESTION 3. THE DES 2009 REPORT ESTABLISHED THRESHOLDS FOR . . . TN concentrations. In this estuary, is TN the correct form of nitrogen on which to focus to address cultural eutrophication? Assuming that the excessive growth of macroalgae and/or epiphytes is one of the primary concerns, what form of nitrogen should be the focus, given detention times in the system? Is the form of nitrogen that should be controlled the same for great bay, the Piscataqua River and Portsmouth Harbor? Based on the available evidence, is it likely that dissolved organic nitrogen is converted to dissolved inorganic nitrogen to a significant degree within this estuary and watershed?

BIERMAN RESPONSE

Yes, TN is the correct form of nitrogen on which to focus to address cultural eutrophication.

I am in agreement with this statement from EPA (2010b):

“Regardless of their source, N and P are present in three main forms: dissolved organic N and P, dissolved inorganic N and P, and particulate N and P (Chapra 1997). These compounds frequently cycle between forms, transforming and reacting between dissolved and particulate fractions. Only dissolved organic and inorganic forms are taken up by microbes and primary producers, and this uptake capacity and rate varies among taxa and environmental conditions.”

“For P, soluble reactive phosphorus (e.g., PO₄) is the form most readily available to plants and algae (Correll 1998). Although soluble PO₄ concentration can be measured directly, it is taken up by plants or converted to other forms quickly in the environment, and measurements of soluble PO₄ may not provide an accurate indication of available P. Therefore, total P (TP) is commonly measured and used as an indicator of the amount of P available to the system. Estimates of P loading have also been combined with lake retention time and P settling rates to model observed chl a concentrations (Vollenweider 1976).”

“For N, inorganic N in the forms of ammonia (NH₃) and nitrate (NO₃) are preferred by plants and algae. Like PO₄, it is often difficult to measure NH₃ and NO₃ frequently enough in most state sampling programs to capture nutrient-plant dynamics. Thus, total N (TN) is commonly used to represent the amount of N in the system and its relationship to primary production.”

For Great Bay Estuary it is not possible to answer the question about the influence of detention times on conversion of nitrogen from unavailable to available forms within the watershed or estuary. To answer this question, a load-response mass balance model would be required that incorporates estuarine hydrodynamics, and nitrogen cycling in the water column and bedded sediments. Such a model does not presently exist for Great Bay Estuary.

DIAZ RESPONSE

The nitrogen cycle is complex with many forms that are biologically active and readily transformed. Processes responsible for the transformation, retention, or removal of nitrogen in shallow coastal systems are diverse and include uptake, release, and mineralization by primary producers and microbes, burial, denitrification, and transport to the coastal ocean.

For managing nutrient driven eutrophication the most important forms are ammonia, nitrate, and nitrite, which when summed represent dissolved inorganic nitrogen (DIN). This is what the DES 2009 Report considers to be DIN. Other fractions considered were dissolved organic nitrogen (DON), nitrogen in phytoplankton, and nitrogen in all other particulate organic matter. The report also states that total nitrogen (TN) is the sum of all dissolved nitrogen plus all particulate nitrogen.

It is generally thought that macroalgae growth is related primarily to DIN loads or concentrations, so a strategy that focused on all forms of DIN might seem appropriate. However, bioavailable compounds in the DON pool, such as amino acids and urea, can make up significant portion of the DON pool and contribute to macroalgal production (Tyler and McGlathery 2006). In addition, labile organic compounds may represent an important source of N for both heterotrophic and autotrophic microorganisms, as well as for benthic plants (Tyler et al. 2001). For the Great Bay system TN is about 38% DIN, about 39% DON, and about 23% PON (from Table 3 in DES 2009 Report). Given that DON and PON can be converted to DIN and taken up directly by macroalgae, a key question with regards to which fractions of nitrogen to control would be: How much of the DON and PON fractions within Great Bay are converted to DIN and how much is taken up directly as DON? Based on the information in the DES 2009 Report, it is not possible to determine the rate of conversion of organic nitrogen to DIN or direct uptake.

Relative to excessive macroalgal production, the timing of availability of DIN and DON would be important as macroalgae occur seasonally. During the times of year that macroalgae are not present, what other species take up the TN fractions?

KENWORTHY RESPONSE

Yes, total nitrogen is the correct and most robust form of nitrogen to use as an indicator of nitrogen status in an estuary. Normally, the transformations of inorganic forms of nitrogen to organic forms and the metabolism of primary producers and microorganisms which process inorganic and nitrogen are extremely rapid. The dynamics are so rapid, and the variability in concentrations so high, standard water quality monitoring programs cannot adequately capture and statistically describe the variability in the inorganic forms of nitrogen (Wazniak et al. 2008, Benson et al. 2013). Unless there was demonstrable evidence that an excessive abundance of recalcitrant forms of organic nitrogen are delivered to the Great Bay estuary, there is no reason to be concerned that measuring total nitrogen is incorrect.

Detention times for the individual segments and the entire system of Great Bay are not quantitatively addressed in the DES 2009 Report, so it is impossible to answer that portion of the question. However, it is well documented that water residence time is an important factor in considering nitrogen loading and eutrophication in coastal systems (Krause Jensen et al. 2008, Latimer and Rego 2010, Benson et al. 2013).

Water residence times and hydrodynamics are important centerpieces of the MEP nitrogen modelling program in Massachusetts and it would be informative for DES to consider incorporating these factors into their assessment.

The information provided in the DES 2009 Report is insufficient for determining if the forms of nitrogen are different in the Great Bay, Piscataqua River and Portsmouth Harbor.

RECKHOW RESPONSE

I think that TN is the best measurement of nitrogen to set as a water quality criterion and for a TMDL to control eutrophication. We know that algae preferentially take up inorganic nitrogen forms (nitrate and ammonium), and over short time scales inorganic nitrogen forms provide the best indicator of near-term future algal growth (if N is limiting). Recently, some scientists have reasoned that since nitrogen is a component of algal cells, then the use of the TN – chlorophyll relationship as a basis for nitrogen control decisions is wrong due to spurious correlation (Lewis and Wurtsbaugh 2008). I think this conclusion is flawed when it is cited as the basis for not using TN to set water quality criterion, since we know that TN reductions can be expected to lead to chlorophyll reductions. In addition, since transformations of nitrogen from one form to another do occur in surface waters, it does not make sense to me to have a (long-term) water quality criterion or TMDL for nitrogen based on any other parameter except TN.

QUESTION 4. THE DES 2009 REPORT WAS PUBLISHED NEARLY FIVE YEARS. . . Ago. To ensure ongoing protection of estuarine resources and water quality based on the latest scientific understandings, the des 2009 report may be updated in the future.

a) If you were charged with updating the DES 2009 Report, what approach would you take given the information now available?

BIERMAN RESPONSE

My answer to this question assumes that the goal of an updated DES 2009 Report is to refine/revise the numeric nutrient criteria based on new data, models and information that has become available since publication of the original document. A caveat to my answer is that improvements in water quality/ecological health in Great Bay Estuary can only be obtained by controlling nutrient loads, not by simply setting numeric nutrient criteria. Such criteria may be beneficial in cases where only narrative criteria exist and progress on nutrient load controls is held hostage to endless arguments over how to translate narrative criteria into quantitative criteria. In my opinion, however, numeric nutrient criteria are a solution to a regulatory problem, not a water quality problem. They are one link in a causal chain that must eventually connect nutrient loads to water quality and/or ecological endpoints in order to develop controls on nutrient loads in the form of TMDLs and/or NPDES permits.

The U.S. EPA itself took this approach in their use of process-based mass balance models by Scavia et al. 2004 and Cerco et al. 2010 to develop nutrient load reduction goals for the Gulf of Mexico (Mississippi River/Gulf of Mexico Watershed Nutrient Task Force, 2008) and the nutrient and sediment TMDLs for Chesapeake Bay (EPA 2010c). These process-based models are load-response models, not empirical stressor-response models, and hence they obviate the need for numeric nutrient criteria because they directly link nutrient loads to response variables that represent water quality impairments (e.g., dissolved oxygen, chlorophyll-a, water clarity and acreage of submerged aquatic vegetation). This reasoning applies not only to process-based mass balance models but can also apply to empirical models. Empirical statistical models were developed for the Gulf of Mexico (Scavia and Donnelly 2007; Turner et al. 2008) and Chesapeake Bay (Hagy et al. 2004). These models are also load-response models and none of them involves numeric nutrient criteria.

To update the DES 2009 Report for the purpose of revising/refining the numeric nutrient criteria, I would use a comprehensive “weight of evidence” approach based on the “triad” of methods discussed above in my response to Question 2a:

- Reference condition approach
- Empirical (statistical) stressor-response analysis
- Process-based (mass balance) models.

I would follow the guidance in EPA (2001) for the reference condition approach, in EPA (2010b) for empirical stressor-response analysis, and in Bierman et al. (2013) for process-based mass balance models.

In following the guidance in EPA (2001), I would conduct a comprehensive and systematic review of site-specific data for other representative marine/estuarine systems in the New England and mid-Atlantic regions. I would place emphasis on spatial classification and segmentation of each system, including Great Bay Estuary, into zones with similar flushing times, bathymetry and sediment physical-chemical characteristics. I would use this information base to develop target thresholds for total nitrogen concentrations for each of the distinct water quality/ecological zones in the Great Bay Estuary. In following the guidance in EPA (2010b), I would place emphasis on fully investigating the influence of the co-varying/confounding variables in my response to Question 1a, and address all of the concerns expressed in my response to Question 1e.

In following the guidance in Bierman et al. (2013), I would select a process-based mass balance model from the Nutrient Modeling Toolbox (NMT) that represents the water quality/ecological endpoints of concern in Great Bay Estuary, and which is compatible with the available data for model set-up, inputs and calibration. There are numerous process-based models that would be appropriate for nutrients, chlorophyll-a and DO, but only a limited number of complex models for submerged aquatic vegetation (eelgrass). A viable alternative would be to use a hybrid approach in which a process-based model would link nutrient loads to chlorophyll-a, DO and underwater light attenuation, and an empirical component would be used to link these process-based outputs to eelgrass. This empirical component would need to be developed using results from the reference condition and empirical stressor-response approaches. See Bierman et al. (2013) for a more complete discussion of hybrid models.

There would be three major benefits to using a process-based mass balance model. First, all of the relevant stressor, response and confounding variables could be represented within the same internally consistent mass balance framework. Second, numeric nutrient criteria could be extracted from load reduction simulations with the calibrated model that achieve the desired water quality/ecological endpoints. Finally, load reduction simulations with the calibrated model could be used directly to develop controls on external loadings in the form of TMDLs and/or NPDES permits.

DIAZ RESPONSE

Given that the DES 2009 Report is five years old, its authors did a good job finding and compiling data for the Great Bay system for nutrients and water quality from January 2000 to December 2008. Any approach to an update would start with a compilation of nutrient and water quality data on Great Bay collected from January 2009 to the present, and integration of new data with previous data compilations. The same data update would be needed for sediments and benthic invertebrate communities.

In parallel with the data updates, DES should evaluate the guidance now available from EPA for applying response-stressor and weight of evidence approaches to setting numerical nutrient criteria. Particular attention needs to be given to the conceptual models used to support regulation of nitrogen.

While it is widely understood that nitrogen is typically limiting in marine systems, it is important to consider phosphorus and develop a two-nutrient control strategy. Recent thinking about restoring the Baltic Sea, and other marine systems, has shifted to a two nutrient solution (Conley et al. 2009). Any

update of the DES 2009 Report should include more detailed assessment of how changes in nutrient ratios will affect the ecosystem.

While the DES 2009 Report is focused on setting numerical criteria for total nitrogen, some consideration needs to be included for what are the sources of nitrogen to the Great Bay system, and which are controllable vs. which are uncontrollable. This becomes important in assessing success of any nitrogen reduction strategy. For example, the sources of nitrogen driving eutrophication in Chesapeake Bay, Narragansett Bay, and northern Gulf of Mexico are different and would require tailored approaches to nutrient reduction (Figure 16, CERN 2010). Basically, are there sufficient pools of controllable nitrogen that can be reduced to meet any set numerical criteria for total nitrogen?

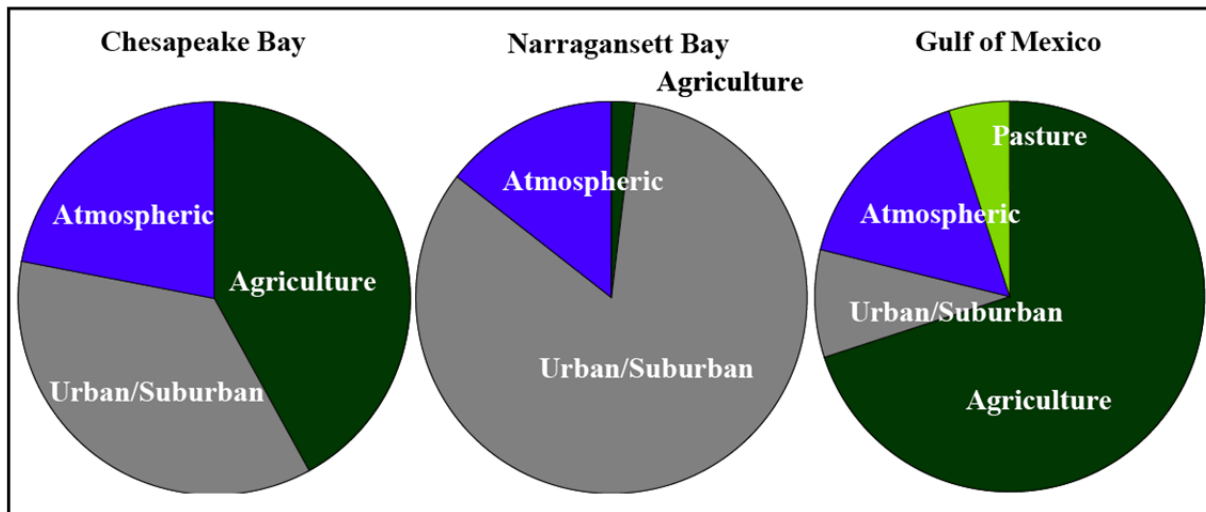


Figure 3. Comparison of the relative contribution of major sources of nitrogen pollution in three coastal ecosystems experiencing hypoxia. Urban/suburban includes both point (industrial and sewage effluent) and nonpoint sources (residential run off). Data sources: Chesapeake Bay: Chesapeake Bay Program; Narragansett Bay: Nixon et al. 2008, Moore et al. 2004; Gulf of Mexico: Alexander et al. 2008.

Figure 16. Variation in sources of nitrogen for three US systems. From CERN (2010). Absent from consideration were oceanic sources, which may be an important sources for systems such as Great Bay.

KENWORTHY RESPONSE

First of all, DES should be complemented for their effort to establish nitrogen criteria in the Great Bay estuary. DES has compiled a complicated, long and very large data set from a wide range of sources to conduct their assessment and to begin addressing nitrogen management and resource protection in the Great Bay estuary. There is compelling scientific information that has identified this problem in many coastal ecosystems, including Great Bay (see citations noted in my responses above). There is also considerable management experience which supports a need for responsible resource agencies and the public and private stakeholders to control nutrient enrichment for the protection of sensitive, productive and valuable estuarine environments. Large scale attention to this problem and the cooperation of scientists and managers, along with public and private financing, has paved a promising path for the protection and restoration of seagrasses in several coastal ecosystems (e.g.; Tomasko et al. 2005, Greening and Janicki 2006, Steward and Green 2007, Orth et al. 2010a; Bensen et al. 2013, Massachusetts Estuaries Project at <https://www.google.com/#q=Massachusetts%20estuaries%20project>).

Along with new and emerging scientific information (locally, nationally and globally) and several comprehensive evaluations of the results of nutrient management programs (see for example Orth et al. 2010 a, b, Greening and Janicki 2006, Tampa Bay National Estuary Program at <http://tbep.tech.org/>), there are also relatively new EPA guidelines (USEPA 2010a, b) recommending more comprehensive approaches to assessing nutrient enrichment and setting criteria. DES was well into their assessment as this new and important scientific information emerged, the recent experiences of other state program were evaluated, and new guidelines were proposed. So it is now possible for DES, the Great Bay estuary stakeholders and the responsible parties in the region to benefit from this new information by a thorough and critical re-evaluation of the approach. The new scientific information, preliminary indications from the applied science, and management experience all strongly suggest there is potential for considerable improvements to be made without sacrificing the data and information already collected and compiled by DES and its' collaborators. The DES 2009 Report would benefit from significantly modifying the approach and collecting new information as per my recommendations discussed below and suggestions made by my fellow panel members.

I would recommend a complete re-evaluation of the updated eelgrass cover data for all of the individual zones in the Great Bay Estuary (see my response to question #1 and Table 1). This re-evaluation would more closely align eelgrass status with the contemporary water quality monitoring data and improve the correspondence analysis between eelgrass, optical water quality, nutrients and environmental conditions in the Great Bay estuary. I would also recommended treating eelgrass status and corresponding water quality data independently for each distinctive zone. This would recognize potential differences in the biophysical characteristics of the zones, as well as the sources of nutrients (loadings), and result in a “zone specific” approach that could be more easily adapted to changing conditions and new information. It is highly likely that this would require the addition of more water quality monitoring stations in each zone as well as a more comprehensive evaluation of the temporal and spatial variability of the different biological, chemical and physical factors affecting the delivery of nutrients to Great Bay, the response of the symptom factors (chlorophyll-a, turbidity, CDOM, K_d) and the corresponding growth, abundance and distribution of eelgrass.

By taking this approach, DES would also benefit by formally addressing eelgrass status in the Great Bay estuary as two different problems. One is a very difficult and uncertain restoration problem in the tributaries that have lost all or most of their eelgrass and the second is a maintenance and conservation problem in the areas where most of the eelgrass resource exists, but is still declining. DES should consider establishing priorities for protecting the largest extent of the resource and adapting the assessment approach to these priorities instead of treating the system with one nitrogen concentration criterion and the expectation that eelgrass will be protected and restored throughout a very complex estuarine ecosystem.

RECKHOW RESPONSE

I would use the methods that I applied in my review of the DES 2009 Report; these methods are described in my responses to Question 1 and Question 2.

b) Would a reference waters approach to establish a TN threshold based on various eutrophic responses such as macroalgae growth, low dissolved oxygen, and eelgrass loss be appropriate and feasible for the great bay estuary? If so, how would you recommend such an approach be developed?

BIERMAN RESPONSE

Yes, but only as one part of a “triad” of methods. See my response to Question 4a.

DIAZ RESPONSE

Approaches that use reference conditions for defining impairment or impact can provide some of the strongest statistical evidence for impairment or impact. These approaches are commonly applied in assessments that use benthic invertebrate communities. When the impact has not yet occurred the designs are known as before-after, control-impact (BACI) study designs. If the impact has occurred, the approach to the assessment changes and requires spatial comparisons between impacted vs. unimpacted sites (Green 1979). Unimpacted sites would provide the control or reference conditions.

There are limitations to a reference waters approach for setting total nitrogen criteria. The first would be the lack of cause-effect between similarities or differences at the reference and impaired sites. Strong stressor-response relationships would still be required and they would have to be applicable to all sites evaluated.

For DES to switch to a reference waters approach to set nitrogen criteria in Great Bay region the following steps need to be followed:

- Identification of impaired waters within the Great Bay region in order to establish the degree of impairment.
- Identification of unimpaired reference waters within the Great Bay region in order to establish reference conditions. If there are no unimpaired waters, can minimally impaired waters be used?
- Given that Great Bay is a relatively small system and watershed (Figure 1), there may be no regions/stations that have sufficient spatial separation for establishing impaired vs. unimpaired waters. Are stations far enough apart but still similar enough for comparison? If this is the case, can unimpaired waters be found outside the Great Bay region that would serve as reference waters?
- Matching of impaired and unimpaired waters to characterize conditions for the primary (Chlorophyll-a and macroalgae) and secondary (benthic invertebrates, sediment quality, DO, and eelgrass) indicators DES will use for setting total nitrogen numeric criteria.
- Establish the stressor-response relationship between total nitrogen and primary and secondary indicators.

If these five points can be adequately addressed, then a reference waters approach may be possible.

KENWORTHY RESPONSE

As per the EPA guidelines and my detailed response to questions #1 and #2, a reference approach should be considered as part of a broader effort that also includes consideration of; 1) nitrogen sources, loading, and hydrodynamic modeling (mass balance modelling), 2) empirical determinations of eelgrass response to selected eutrophication stressors, and 3) empirically ruling out other factors which affect eelgrass growth, abundance and distribution in the Great Bay estuary. Most importantly, DES **should not** consider one reference site for the entire Great Bay estuary. But rather, each zone, or the aggregation of a few zones, should have their own reference condition established depending on the bio-physical similarities and dissimilarities between zones.

RECKHOW RESPONSE

I know that the USEPA has promoted the reference waters approach. I am not a proponent of this strategy as the single basis for setting a water quality criterion. If a reference waters approach is to be used as a basis for TN criteria for the Great Bay, it should be considered as only one of several “lines of evidence” that are needed.

c) Are there other approaches that you would recommend as alternatives for setting site specific nutrient criteria for the tidal Piscataqua and Cocheco rivers?

BIERMAN RESPONSE

The “triad” approach recommended in my response to Question 4a would also be appropriate for the tidal Piscataqua and Cocheco Rivers. To the extent that these assessment zones are more strongly influenced by different physical factors (e.g., flushing rates) than portions of the Great Bay Estuary located further downstream, the process-based mass balance modeling approach might provide a stronger line of evidence than the reference condition or empirical stressor-response approaches.

DIAZ RESPONSE

The approaches followed in the DES 2009 Report for setting total nitrogen concentration criteria rely on stressor-response and weight of evidence methodologies framed and guided by two conceptual models of nutrient enrichment and eutrophication (see Question 1 response). Other approaches would involve numerical modeling or shifting to total nitrogen load as the criteria.

KENWORTHY RESPONSE

According to the DES 2008 report, *“Eelgrass is not known to have been present in the Cocheco River. The historic sources did not map and current eelgrass maps do not show eelgrass in this zone. Available chlorophyll-a data indicate compliance with the chlorophyll-a criterion. Since there are no chlorophyll-a impairments in this zone, an impairment for nutrients per Env 1703.14 is not justified.”* Based on this assessment, I would recommend that the Cocheco River be treated very differently from the tidal Piscataqua which still has eelgrass present, but declining. With respect to eelgrass, it has never been documented in the Cocheco, so it would be irresponsible to set criteria for eelgrass based on any other segments of the Great Bay estuary.

RECKHOW RESPONSE

I would use the methods that I applied in my review of the DES 2009 Report; these methods are described in my responses to Question 1 and Question 2.

d) Do you have any recommendations for the long-term (10-year) monitoring and evaluation of the estuary to assess changes in conditions over time?

BIERMAN RESPONSE

Long-term monitoring and evaluation of the estuary should be conducted within the larger context of an overall decision support system. An adaptive management framework should be used for this decision support system, and should be a framework for integrating continued monitoring, data analysis and process-based mass balance model to improve scientific understanding and reduce uncertainties. A relevant example would be the recommendations in the Massachusetts Estuary Project (MEP) Linked Watershed Embayment Model Peer Review (Scientific Peer Review Panel 2011).

DIAZ RESPONSE

Irrespective of which set of methods DES follows in setting limits on total nitrogen, additional data are needed to link total nitrogen to total ecosystem functioning of the Great Bay system. Before launching into data collection, consider that it is well-established dogma that estuarine ecosystems are complex and are driven by a complex combination of top-down, bottom-up, internal, and external factors. Cloern (2001) captures much of this complexity in Figure 17. Basically, there are no simple cause-effect relationships, it is all interactions. Therefore to focus limited resources on what is essential for setting nitrogen criteria within Great Bay, a detailed conceptual model of all sources of nitrogen entering Great Bay and interactions of ecosystem components with nitrogen would be needed. Evaluation of data gaps within this overall model framework combined with best professional judgment will guide both which linkages are most important, and which short-term and long-term datasets are needed.

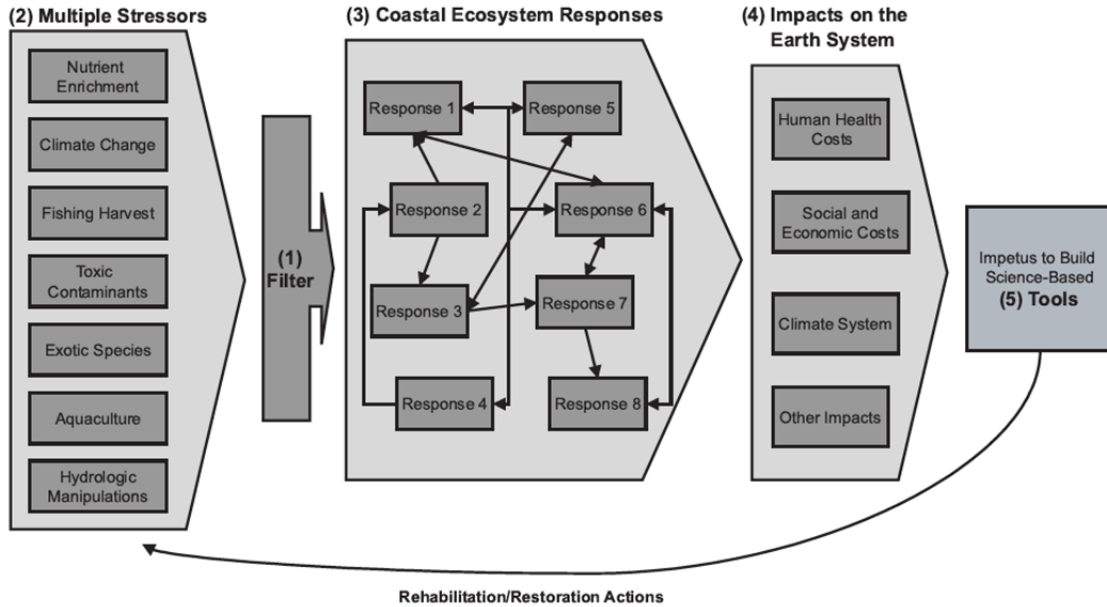


Fig. 24. One view of the next (Phase III) conceptual model of coastal eutrophication, organized around 5 basic questions concerning: (1) the system attributes that act as a filter to modulate the responses to nutrient enrichment; (2) nutrient enrichment as 1 of many interacting stressors; (3) the complex linkages between responses to multiple stressors; (4) impacts of change in coastal ecosystems on the Earth system, including aspects that influence sustainability of the human population; and (5) the application of a deeper and broader scientific understanding of coastal eutrophication to produce a set of tools for building rational management strategies and action plans for ecosystem rehabilitation/restoration

Figure 17. Conceptual model for complexity of interactions within estuarine systems.

From Cloern (2001).

KENWORTHY RESPONSE

I would urge DES to follow the recommendations I have suggested in my responses above to questions #1, 2 and 3 and the specific recommendations made by the other members of this review panel. To summarize the most important points I will briefly re-iterate that DES should: 1) incorporate the more recent eelgrass data (Table 1) into their assessment and align these data more closely in time and space with a more rigorous analysis of the water quality and light attenuation data; 2) follow the most recent guidelines by EPA and its' expert panel reviews which recommend a broader approach to the assessment process by incorporating stressor response analyses, appropriate reference conditions, and process based modelling; 3) consider addressing the different zones in the Great Bay estuary independently for the assessment of eelgrass, water quality status, and reference condition; 4) improve the assessment by quantitatively recognizing and treating the status of eelgrass and eutrophication in the different zones as either a conservation and maintenance problem or a restoration problem; 5) incorporate more basic information and understanding of eelgrass biology (e.g., reproductive biology) and ecology as it pertains to eutrophication, eelgrass loss, and eelgrass recovery; and 6) review and evaluate the more recent basic and applied scientific literature cited in this review to gain a better understanding of the problem in Great Bay and refinements in the assessment process.

In addition to these specific recommendations, I also suggest that DES consider using a properly calibrated bio-optical water quality model to assess the effects of chlorophyll-a, turbidity, and CDOM on

light attenuation (K_d) and eelgrass abundance and distribution (Gallegos 2001, Gallegos 2005, Gallegos and Kenworthy 1996, Biber et al. 2008, Kenworthy et al. 2013). As indicated earlier, the data used by DES from the Morrison et al. (2007) study show that all three monitored optical water quality components (chlorophyll-a, turbidity, CDOM) are contributing to light attenuation in the Great Bay estuary. Based on the 2009 assessment report, it appears that DES has undervalued the contributions of turbidity and CDOM and placed a disproportionate emphasis on chlorophyll-a. Distinguishing the relative importance or the effects of each one of these variables using linear regression (simple or multiple) analyses can be significantly improved by using a bio-optical water quality model to calculate K_d and having some knowledge about eelgrass light requirements (Gallegos 2001, Kenworthy et al. 2013). The bio-optical modelling approach recognizes that K_d is an “apparent” optical property and can be calculated by using inherent optical properties (absorption and scattering) which are additive and directly related to concentrations of regularly measured water quality parameters (chlorophyll-a, turbidity, CDOM).

A properly calibrated model can be used to directly evaluate the sensitivity of individual water quality parameters as well as the combination of parameters that are affecting K_d (see Figure 10 in Gallegos 2001 and Figures 6 and 7 in Kenworthy et al. 2013) and ultimately influencing the depth distribution and abundance of eelgrass. A bio-optical model recently calibrated in Massachusetts (Kenworthy et al. 2013) could be easily transferred for application to existing and newly collected optical water quality data in the Great Bay estuary with a minor amount of effort to calibrate the model. This model could be used to quantitatively calculate eelgrass restoration depths in each of the designated zones of the Great Bay estuary.

RECKHOW RESPONSE

My analysis indicates that the available NH DES data provide a weak basis for setting TN criteria for the Great Bay Estuary. If you are making decisions that have substantial economic and societal consequences, then you want to be confident in your decision. I think that too often we spend too little on planning that informs decisions that have major consequences. To remedy this, I recommend that NH DES invest wisely in future water quality monitoring, assessment, and modeling.

I can go on at length as to how you might do this, but in brief, you first need to establish a water quality monitoring program that is not based on convenience sampling, but rather is focused on the major uncertainties, given your objectives to set numerical nitrogen criteria. While to some degree we know the answers, the key questions remain: (1) what are the designated uses that you are trying to protect? (2) What criteria are the best measurable indicators of attainment of the designated uses? (3) Over what space/time scales must you measure these criteria to achieve an acceptable error level for the determination of attainment/nonattainment? Answers to these questions, perhaps utilizing a water quality model, should provide the basis for a multi-year monitoring program that should provide more confidence in your decision on TN criteria.

REFERENCES

- Benson, J.L., D. Schlezinger and B.L. Howes. 2013. Relationship between nitrogen concentration, light and *Zostera marina* habitat quality and survival in southeastern Massachusetts estuaries. *Journal of Environmental Management*. 131:129-137.
- Biber, P., Gallegos, C, Kenworthy, W. J. 2008. Calibration of a bio-optical model in the North River, North Carolina (Albemarle-Pamlico Sound): A tool to evaluate water quality impact on seagrasses. *Estuaries and Coasts* 31:177-191.
- Bierman, V.J., Jr., J.V. DePinto, D.W. Dilks, P.E. Moskus, T.A.D. Slawecki, C.F. Bell, S.C. Chapra and K.F. Flynn. 2013. Modeling Guidance for Developing Site-Specific Nutrient Goals. Water Environment Research Foundation, Alexandria, Virginia.
- Bricker, S.B., C.G. Clement, D.E. Pirhalla, S.P. Orlando, and D.R.G. Farrow. 1999. National Estuarine Eutrophication Assessment: Effects of Nutrient Enrichment in the Nation's Estuaries. NOAA, National Ocean Service, Special Projects Office and the National Centers for Coastal Ocean Science. Silver Spring, MD. 71 pp.
- Bricker, S., B. Longstaff, W. Dennison, A. Jones, K. Boicourt, C. Wicks, and J. Woerner. 2007. Effects of Nutrient Enrichment In the Nation's Estuaries: A Decade of Change. NOAA Coastal Ocean Program Decision Analysis Series No. 26. National Centers for Coastal Ocean Science, Silver Spring, MD. 328 pp.
- Bricker, S.B., B. Longstaff, W. Dennison, A. Jones, K. Boicourt, C. Wicks, and J. Woerner. 2008. Effects of nutrient enrichment in the nation's estuaries: A decade of change. *Harmful Algae* 8:21–32.
- Burkholder, J.M., D.A. Tomasko and B.W. Touchette. 2007. Seagrasses and eutrophication. *Journal of Experimental Marine Biology and Ecology* 350:46-72.
- Cerco, C.F., S. Kim and M.R. Noel. 2010. The 2010 Chesapeake Bay Eutrophication Model. Report to the U.S. Environmental Protection Agency Chesapeake Bay Program and to the U.S. Army Engineer Baltimore District. U.S. Army Engineer Research and Development Center, Vicksburg, MS.
- Christman, C.S. and D.M. Dauer. 2003. An approach for identifying the causes of benthic degradation in Chesapeake Bay. *Environmental Monitoring and Assessment* 81:187–197.
- Cloern, J.E. 2001. Our evolving conceptual model of the coastal eutrophication problem. *Marine Ecology Progress Series*. 210:223-253.
- Committee on Environment and Natural Resources. 2010. Scientific Assessment of Hypoxia in U.S. Coastal Waters. Interagency Working Group on Harmful Algal Blooms, Hypoxia, and Human Health of the Joint Subcommittee on Ocean Science and Technology. Washington, DC.
- Conley, D.J., H.W. Paerl, R.W. Howarth, D.F. Boesch, S.P. Seitzinger, K.E. Havens, C. Lancelot, and G.E. Likens. 2009. Controlling eutrophication by reducing both nitrogen and phosphorus. *Science* 323:1014-1015.
- Costello, C., Kenworthy, W.J. 2011. Twelve-year mapping and change analysis of eelgrass (*Zostera marina*) areal abundance in Massachusetts (USA) identifies Statewide Declines. *Estuaries and Coasts*. 34:232-242.
- Dauer, D.M., Ranasinghe, J.A. and Rodi, A.J. 1992. Effects of low dissolved oxygen levels on the macrobenthos of the lower Chesapeake Bay. *Estuaries* 15:384–391.
- Dennison, W.C., and R.S. Alberte. 1982. Photosynthetic responses of *Zostera marina* L. (eelgrass) to in situ manipulations of light intensity. *Oecologia* 55:147-144.

- Dennison, W.C. and R.S. Alberte. 1985. Role of daily light period in the depth distribution of *Zostera marina* (eelgrass). *Marine Ecology Progress Series* 25, 51-61.
- Dennison, W.C. and R.S. Alberte. 1986. Photoadaptation and growth of *Zostera marina* L. (eelgrass) transplants along a depth gradient. *Journal of Experimental Biology and Ecology* 98, 265-282.
- Dennison, W.J., Orth, R.J., Moore, K.A., Stevenson, J.C., Carter, V., Kollar, S., Bergstrom, P.W., Batiuk, A., 1993. Assessing water quality with submerged aquatic vegetation. *BioScience* 43, 86-94.
- Diaz, R.J. and R. Rosenberg. 1995 Marine benthic hypoxia: a review of its ecological effects and the behavioural responses of benthic macrofauna. *Oceanogr. Mar. Biol. Ann. Rev.* 33:245-303.
- Diaz, R.J. and D.L. Breitburg. 2009. The Hypoxic Environment. In J.G. Richards, A.P. Farrell, and C.J. Brauner, eds: *Fish Physiology*, Vol 27, Burlington: Academic Press, pp.1-23.
- Duarte, C. M., Marbà, N., Krause-Jensen D., Sánchez-Camacho, M., 2007. Testing the predictive power of seagrass depth limit models. *Estuaries and Coasts* 30:652-656.
- Frederiksen, M., D. Krause-Jensen, M. Holmer and J.S. Laursen. 2004a. Long-term changes in area distribution of eelgrass (*Zostera marina*) in Danish coastal waters. *Aquatic Botany* 167-181.
- Frederiksen, M., D. Krause-Jensen, M. Holmer and J.S. Laursen. 2004b. Spatial and temporal variation in eelgrass (*Zostera marina*) landscapes: influence of physical setting. *Aquatic Botany*. 78:147-165.
- Gallegos, C.L. 2001. Calculating optical water quality targets to restore and protect submersed aquatic vegetation: overcoming problems in partitioning the diffuse attenuation coefficient for photosynthetically active radiation. *Estuaries* 24, 381-397.
- Gallegos, C. L. 2005. Optical water quality of a blackwater river estuary: the Lower St. Johns River, Florida, USA. *Estuarine Coastal and Shelf Science* 63, 57-72.
- Gallegos, C.L. and W.J. Kenworthy. 1996. Seagrass depth limits in the Indian River Lagoon (Florida, U.S.A.): Application of an optical water quality model. *Estuarine Coastal Shelf Science*. 42:267-288
- Green, R.H. 1979. *Sampling design and statistical methods for environmental biologists*, Wiley, Chichester.
- Greening, H and A. Janicki. 2006. Toward reversal of eutrophic conditions in a subtropical estuary: water quality and seagrass response to nutrient loading reductions in Tampa Bay, FL, USA. *Environmental Management* 38:163-178.
- Hagy, J.D., W.R. Boynton, C.W. Keefe and K.V. Wood. 2004. Hypoxia in Chesapeake Bay, 1950-2001: Long-term change in relation to nutrient loading and river flow. *Estuaries* 27(4):634-658.
- Hauxwell, J., Cebrian, J., Furlong, C., Valiela, I., 2001. Macroalgal canopies contribute to eelgrass (*Zostera marina*) decline in temperate estuarine ecosystems. *Ecology* 82, 1007-1022.
- Hauxwell, J., Cebrian, J., Valiela, I., 2003. Eelgrass *Zostera marina* loss in temperate estuaries: relationship to land-derived nitrogen loads and effect of light limitation imposed by algae. *Marine Ecology Progress Series*, 247, 59-73.
- Holmer, M. and E.J. Bondgaard. 2001. Photosynthetic and growth response of eelgrass to low oxygen and high sulfide concentrations during hypoxic events. *Aquatic Botany* 70:29-38.
- Hyland, J., L. Balthis, I. Karakassis, P. Magni, A. Petrov, J. Shine, O. Vestergaards, and R. Warwick. 2005. Organic carbon content of sediments as an indicator of stress in the marine benthos. *Marine Ecology Progress Series*. 295:91-103.
- Kemp, W.M., Batiuk, R., Bartleson, R., Bergstrom, P., Carter, V., Gallegos, C. L., Hunley, W., Karrh, L., Koch, E. W., Landwehr, J. M., Moore, K. A., Murray, L., Naylor, M., Rybicki, N.B., Stevenson, J.

- C., Wilcox, D. J. , 2004. Habitat requirements for submerged aquatic vegetation in Chesapeake Bay: water quality, light regime and physical-chemical factors. *Estuaries* 27: 363-377.
- Kenworthy, W.J. and M.S. Fonseca. 1996. Light Requirements of seagrasses *Halodule wrightii* and *Syringodium filiforme* derived from the relationship between diffuse light attenuation and maximum depth distribution. *Estuaries* 19:740-750.
- Kenworthy, W.J., C.L. Gallegos, C. Costello, D. Field and G. Di Carlo. 2013. Dependence of eelgrass (*Zostera marina*) light requirements on sediment organic matter in Massachusetts coastal bays: Implications for remediation and restoration. *Mar. Pollut. Bull.* (2013), <http://dx.doi.org/10.1016/j.marpolbul.2013.11.006>.
- Koch, E. 2001. Beyond light: Physical, geological and geochemical parameters as possible submersed aquatic vegetation habitat requirements. *Estuaries*. 24(1):1-17.
- Krause-Jensen, D., Sagert, S., Schubert, H., Boström, C. 2008. Empirical relationships linking distribution and abundance of marine vegetation to eutrophication. *Ecological Indicators* 8, 515-529.
- Larkum, A.W.D., R.J. Orth and C.M. Duarte (eds.). 2006. *Seagrasses: Biology, Ecology and Conservation*. Springer, Dordrecht, The Netherlands. 691pp.
- Latimer, J.S., Rego, S.A., 2010. Empirical relationship between eelgrass extent and predicted watershed-derived nitrogen loading for shallow New England estuaries *Estuarine, Coastal and Shelf Science* 90, 231-240.
- Lewis, W. M.; Wurtsbaugh, W. A. Control of lacustrine phytoplankton by nutrients: Erosion of the phosphorus paradigm. *Int. Rev. Hydrobiol.* 2008, 93, 446–465.
- McGlathery, K.J., Sundbäck, K., Anderson, I.C., 2007. Eutrophication in shallow coastal bays and lagoons: the role of plants in the coastal filter. *Marine Ecology Progress Series* 348, 1-18
- Mississippi River/Gulf of Mexico Watershed Nutrient Task Force. 2008. *Gulf Hypoxia Action Plan 2008 for Reducing, Mitigating, and Controlling Hypoxia in the Northern Gulf of Mexico and Improving Water Quality in the Mississippi River Basin*. Washington, DC.
- Moksnes, P.O., M. Gullström, K. Tryman and S. Baden. 2008. Trophic cascades in a temperate seagrass community. *Oikos* 117:763-777.
- Moore, K.A. and F.T. Short. 2006. *Zostera: Biology, Ecology and Management*. Pp 3 61-386 in: Larkum, A.W.D., R.J. Orth and C.M. Duarte (eds.), *Seagrasses: Biology, Ecology and Conservation*. Springer, Dordrecht, The Netherlands.
- Morrison, J.R., T.K. Gregory, S. Pe'eri, W. McDowell and P. Trowbridge. 2008. *Using Moored Arrays and Hyperspectral Aerial Imagery to Develop Nutrient Criteria for New Hampshire's Estuaries*. Final Report to The New Hampshire Estuaries Project.
- Neckles, H.L., Wetzel, R.L. and Orth, R.J. 1993. Relative effects of nutrient enrichment and grazing on epiphyte-macrophyte (*Zostera marina* L.) dynamics. *Oecologia* 93:285–295.
- Orth, R. J., M. R. Williams, S. R. Marion, D. J. Wilcox, T. J. B. Carruthers, K. A. Moore, W. M. Kemp, W. C. Dennison, N. Rybicki, P. Bergstrom, and R. A. Batiuk. 2010a. Long term trends in submersed aquatic vegetation (SAV) in Chesapeake Bay, USA, related to water quality. *Estuaries and Coasts* 33:1144-1163. DOI 10.1007/s12237-010-9311-4.
- Orth, R.J., S.R. Marion, K.A. Moore, D. J. Wilcox. 2010. Eelgrass (*Zostera marina* L.) in the Chesapeake Bay Region of Mid-Atlantic Coast of the USA: Challenges in Conservation and Restoration. *Estuaries and Coasts* 33:139-150. DOI 10.1007/s12237-009-9234-0.

- Piscataqua Region Estuaries Partnership. 2013. State of Our Estuaries. University of New Hampshire, Durham, NH.
- Ralf, P.J., Durako, M.J., S. Enriquez, S., Collier, C.J., Doblin, M.A., 2007. Impact of light limitation on seagrasses. *Journal of Experimental Marine Biology and Ecology* 350, 176-193.
- Rosenberg, R., Blomqvist, M., Nilsson, H.C., Cederwall, H., and Dimming, A. 2004. Marine quality assessment by use of benthic species-abundance distributions: a proposed new protocol with in the European Union Water Framework Directive. *Marine Pollution Bulletin* 49:728–739.
- Scavia, D., D. Justić, and V.J. Bierman, Jr. 2004. Reducing hypoxia in the Gulf of Mexico: advice from three models. *Estuaries*. 27:419-425.
- Scavia, D. and K.A. Donnelly. 2007. Reassessing hypoxia forecasts for the Gulf of Mexico. *Environmental Science and Technology*. 41:8111-8117.
- Scientific Peer Review Panel. 2011. Massachusetts Estuary Project (MEP) Linked Watershed Embayment Model Peer Review. Prepared for Barnstable County, Massachusetts. Cape Cod Water Protection Collaborative.
- Short, F.T. 2008. Eelgrass distribution in the Great Bay Estuary for 2007. A Final Report to the Piscataqua Region Estuaries Partnership, University of New Hampshire Durham, NH. Published online at http://www.nhep.unh.edu/resources/pdf/eelgrass_distribution_in-unh-08b.pdf
- Short, F.T., Burdick, D.M. 1995. Mesocosm experiments quantify the effects of eutrophication on eelgrass, *Zostera marina*. *Limnology and Oceanography* 40, 740-749.
- Short, F.T., Burdick, D.M., 1996. Quantifying eelgrass habitat loss in relation to housing development and nitrogen loading in Waquoit Bay, MA. *Estuaries* 19:730-739.
- Short, F.T. and S. Wyllie Echeverria. 1996. Natural and human induced disturbance of seagrasses. *Environmental Conservation*. 23:17-27.
- Short, F.T., R.C. Davis, B.S. Koop, C.A. Short and D.M. Burdick. 2002. Site selection model for optimal restoration of eelgrass, *Zostera marina* L. *Marine Ecology Progress Series* 227:253-267.
- Short, F.T., A.S. Klein, D.M. Burdick and G.E. Moore. 2012. The eelgrass resource of southern New England and New York: Science in support of management and restoration success. Final Report to the NOAA Restoration Center Community-based Restoration Program (CRP). 93pp. <http://www.conservationgateway.org/Files/Pages/eelgrass-resource-souther.aspx>
- Steward, J.S. and W.C. Green. 2007. Setting load limits for nutrients and suspended solids based upon seagrass depth-limit targets. *Estuaries and Coasts* 30:657-670.
- Taylor, D.I., S.W. Nixon, S.L. Granger, B.A. Buckley, J.P. McMahon, H.-J. Lin. 1995. Responses of coastal lagoon plant communities to different forms of nutrient enrichment-a mesocosm experiment. *Aquatic Botany* 52:19-34.
- Thayer, Gordon W., W. Judson Kenworthy, and Mark S. Fonseca. 1984. The ecology of eelgrass meadows of the Atlantic coast: a community profile. U.S. Fish and Wildl. Serv. FWS/OBS-84/02. 147pp.
- Tomasko, D.A., C.A. Corbett, H.S. Greening and G.E. Raulerson. 2005. Spatial and temporal variation in seagrass coverage in southwest Florida: assessing the relative effects of anthropogenic nutrient load reductions and rainfall in four contiguous estuaries. *Marine Pollution Bulletin* 50:797-805.
- Turner, R.E., N.N. Rabalais and D. Justić. 2008. Gulf of Mexico hypoxia: alternate states and a legacy. *Environmental Science and Technology*. 42:2323-2327.

- Tyler, A.C., McGlathery, K.J. and Anderson, I.C. 2001. Macroalgal mediation of dissolved organic nitrogen fluxes in a temperate coastal lagoon. *Estuarine Coastal Shelf Science* 53:155–68.
- Tyler, A.C. and K.J. McGlathery. 2006. Uptake and release of nitrogen by the macroalgae *Gracilaria vermiculophylla* (Rhodophyta). *Journal of Phycology* 42:515–525.
- U.S. Environmental Protection Agency. 2001. Nutrient Criteria Technical Guidance Manual: Estuarine and Coastal Marine Waters. EPA-822-B-01-03. Office of Water. Washington, DC.
- U.S. Environmental Protection Agency. 2008. National Coastal Condition Report III. EPA-842-R-08-002. U.S. Environmental Protection Agency, Office of Water and Office of Research and Development, Washington, DC.
- U.S. Environmental Protection Agency. 2009. Empirical Approaches for Nutrient Criteria Derivation. Science Advisory Board Review Draft. Office of Science and Technology, Office of Water. Washington, DC.
- U.S. Environmental Protection Agency. 2010a. Review of Empirical Approaches for Nutrient Criteria Derivation. Science Advisory Board, Office of the Administrator. Washington, DC.
- U.S. Environmental Protection Agency. 2010b. Using Stressor-response Relationships to Derive Numeric Nutrient Criteria. EPA-820-S-10-001. Office of Science and Technology, Office of Water. Washington, DC.
- U.S. Environmental Protection Agency. 2010c. Chesapeake Bay Total Maximum Daily Load for Nitrogen, Phosphorus and Sediment. Region 3, Philadelphia, PA, and Annapolis, MD, Region 2, New York, NY, Office of Water, Washington, DC.
- U.S. Environmental Protection Agency. 2011. Endocrine disruptor screening program. Weight-of-evidence: Evaluating results of EDSP Tier 1 screening to identify the need for Tier 2 testing. Office of Chemical Safety and Pollution Prevention, US Environmental Protection Agency, Washington DC. 47 p.
- U.S. Environmental Protection Agency. 2012. National Coastal Condition Report IV. EPA-842-R-10-003. U.S. Environmental Protection Agency, Office of Water and Office of Research and Development, Washington, DC.
- Valiela, I., Foreman, K., LaMontague, M., Hersh, D., Costa, J., Peckol, P., DeMeo-Anderson, B., D'Avanzo, C., Babione, M., Sham, C.H., Brawley, J., Lajth, K., 1992. Coupling of watersheds and coastal waters: sources and consequences of nutrient enrichment in Waquoit Bay, Massachusetts. *Estuaries* 15, 443-457. Vaudry et al. 2010.
- Vaudrey, J.M.P., Kremer, J.N., Branco, B.F., Short, F.T. 2010. Eelgrass recovery after nutrient enrichment reversal. *Aquatic Botany* 93, 237-243.
- Vaquar-Sonyer, R. and Duarte, C.M. 2008. Thresholds of hypoxia for marine biodiversity. *Proceedings of the National Academy of Science* 105:15452-15457.
- Waycott, M., C.M. Duarte, T.J.B. Carruthers, R.G. Orth, W.C. Dennison, S. Olyarnik, A. Calladine, J.W. Fourqurean, K.A. Heck, Jr., A.R. Hughes, G.A. Kendrick, W. J. Kenworthy, F.T. Short, and S.L. Williams. 2009. Accelerating loss of seagrasses across the globe threatens coastal ecosystems. *Proceedings of the National Academy of Sciences* 106:12377-12381.
- Wazniak, C.E., M.R. Hall, T.J.B. Carruthers, B. Sturgis, W.C. Dennison, and R.J. Orth. 2007. Linking water quality to living resources in a mid-Atlantic lagoon system, USA. *Ecological Applications* 17: S64–S78.

Whitledge, T.E. 1985. Nationwide review of oxygen depletion and eutrophication in estuarine and coastal waters: northeast region. Report to U.S. Dept. of Commerce, NOAA, National Ocean Service. Rockville, MD. 718 p.