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Eelgrass *Zostera marina* loss in temperate estuaries: relationship to land-derived nitrogen loads and effect of light limitation imposed by algae

Jennifer Hauxwell[1,2,4,*], Just Cebrián[1,3,5], Ivan Valiela[1]

[1]Boston University Marine Program, Marine Biological Laboratory, Woods Hole, Massachusetts 02543, USA
[2]Wisconsin Department of Natural Resources, DNR Research Center, 1350 Femrite Drive, Monona, Wisconsin 53716, USA
[3]Dauphin Island Sea Lab, 101 Bienville Boulevard, PO Box 369-370, Dauphin Island, Alabama 36528, USA

[4]*Present address:* Wisconsin Department of Natural Resources, DNR Research Center, 1350 Femrite Drive, Monona, Wisconsin 53716, USA

[5]*Present address:* Dauphin Island Sea Lab, 101 Bienville Boulevard, PO Box 369-370, Dauphin Island, Alabama 36528, USA

ABSTRACT: In this paper, we explicitly link changes in community structure of estuarine primary producers to measured nitrogen loading rates from watersheds to estuaries, and quantify the relationship between nitrogen load, annual dynamics of algal growth and *Zostera marina* L. productivity, and overall eelgrass decline at the watershed-estuarine scale in estuaries of Waquoit Bay, Massachusetts, USA. Substantial eelgrass loss (80 to 96% of bed area lost in the last decade) was found at loads of ~30 kg N ha^{-1} yr^{-1}, and total disappearance at loads ≥60 kg N ha^{-1} yr^{-1}. Rather than decreased eelgrass growth rates, we observed an exponential decrease in shoot densities and bed area (and subsequently, areal production) as nitrogen loads increased, suggesting that eelgrass decline in higher-nitrogen estuaries of the Waquoit system occurred largely via lack of recruitment or enhanced mortality of established shoots. Similar to the patterns observed in many other systems and the experimental results obtained in laboratories or mesocosms, the relationship we observed between nitrogen loads and eelgrass health within the Waquoit system was indirect: increased nitrogen stimulated growth and standing stocks of algal producers, that may have caused severe light limitation of eelgrass. From light budgets that considered water column, epiphyte, and macroalgal shading, we estimated chronic, severe light limitation to newly recruiting shoots in higher-nitrogen estuaries, due mainly to shading by a coexisting ≤15 cm macroalgal canopy. Two management recommendations aimed at eelgrass preservation emerge from this work. First, development and management of watersheds must be conducted such that land-derived nitrogen loading to estuaries is restricted. In the Waquoit Bay estuaries, for example, eelgrass is absent or rapidly disappearing from all but those receiving the lowest (≤15th percentile) loads. Second, shoot density and meadow area, rather than growth rates per shoot, seem to be adequate variables for routine monitoring of eelgrass health. We also show that the shift from eelgrass- to algae-dominated communities has important consequences for total system primary production and carbon and nitrogen cycling. Estimated total primary production by coastal assemblages in the Waquoit Bay system was 135% higher in estuaries receiving relatively high versus low loads of land-derived nitrogen, suggesting important trophic and biogeochemical alterations to temperate estuarine ecosystems as a result of eutrophication.

KEY WORDS: Seagrass · Macroalgae · Epiphytes · Phytoplankton · Irradiance · Waquoit Bay · Eutrophication · Estuary

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INTRODUCTION

Eelgrass *Zostera marina* L. habitat has been lost from temperate estuaries worldwide. This loss has occurred via several mechanisms, both natural and human-induced. Natural disturbances, such as hurricanes, earthquakes, ice scour, bioturbation, and herbivory, may account for a small percentage of worldwide loss (Short & Wyllie-Echeverria 1996). Wasting disease caused an extensive die-off in the 1930s along the

*Email: jennifer.hauxwell@dnr.state.wi.us

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Atlantic coastlines of North America (Cottam 1933, Cotton 1933, Renn 1935) and Denmark (Petersen 1934, Rasmussen 1973), but is now only locally important (Short et al. 1986). Human-induced disturbances, such as dredging, addition of docks, mooring of boats, harvesting of shellfish using rakes or trawls, and use of motorboats in shallow waters have created 'scarred' areas within eelgrass meadows. Sediment transport and herbicide runoff as a result of development and agricultural activities in adjoining land parcels may also have affected eelgrass habitat (Kemp et al. 1983).

Anthropogenic nitrogen loading from watersheds to estuaries, or increased delivery of nutrients into receiving waters generated by human activities, may also be an important cause of eelgrass loss (Short et al. 1995, Valiela et al. 2000b, Cloern 2001, Hauxwell et al. 2001). Increased nutrient loads into estuarine waters result from the disproportionate increase in human population near the coast (as compared to inland populations) and associated transformation of natural land into urban development, agricultural land, and recreational facilities (i.e. golf courses: Nixon 1995, Cloern 2001). Recent estimates reveal that 40% of the world's population live within 100 km of the coastline (Cohen et al. 1997), and it is predicted that this imbalance will become greater, because coastal populations have faster growth rates than inland populations. In fact, anthropogenic nitrogen loading is now viewed as one of the most pervasive, world-wide, human impacts on estuaries (NRC 1994, Jackson et al. 2000, Tilman et al. 2001).

Increasing evidence shows that an important proximate cause by which increased nitrogen supply leads to eelgrass decline is via intense light limitation due to the overgrowth of fast-growing, nitrogen-limited algal producers (Kemp et al. 1983, Short et al. 1993, 1995, Short & Wyllie-Echeverria 1996, Valiela et al. 1997b, Hauxwell et al. 2001). Increased nitrogen delivery into estuarine waters stimulates the growth of opportunistic algae, including phytoplankton, epiphytes, and filamentous macroalgae (Sand-Jensen & Borum 1991, Duarte 1995, Short et al. 1995, Taylor et al. 1995b, Hauxwell et al. 1998), which may attenuate a large percentage of light that was available to eelgrass under low-nitrogen loads. Other processes associated with large accumulations of algal producers, such as anoxia (Pregnall et al. 1984, Koch et al. 1990), redox changes resulting from low-oxygen concentration (i.e. high-sulfide concentrations: Goodman et al. 1995, Terrados et al. 1999), and high and toxic ammonium concentrations (Van Katwijk et al. 1997), may also contribute to eelgrass decline. Direct effects of nitrogen loading, such as high and toxic concentrations of nitrate have also been found to cause eelgrass decline (Burkholder et al. 1992, 1994). In aggregate, however, the available literature indicates that most often, light limitation is a primary mechanism of eelgrass decline under enhanced eutrophication (see reviews by Duarte 1995 and Cloern 2001).

The associations between increased nitrogen loading, light interception by algal producers, and seagrass decline have been largely established with experimental manipulations in laboratory microcosms and mesocosms (Twilley et al. 1985, Burkholder et al. 1994, Neckles et al. 1994, Short et al. 1995, Taylor et al. 1995a,b, Moore & Wetzel 2000). Large-scale comparisons, integrating changes in nitrogen delivery, algal growth, and eelgrass loss at the watershed–estuarine level are lacking. Development of numerical relationships between nitrogen loading and response of estuarine producers at the watershed–estuary scale is important for 3 reasons. First, it is necessary for understanding how interactions among nutrients, algae, and seagrasses observed under laboratory conditions translate into natural, large-scale scenarios. Second, it can assist in developing efficient management practices for eelgrass preservation. For instance, knowledge of the threshold land-derived nitrogen loading rate at which eelgrass declines, and understanding the proximate causes for such decline, would help in formulating policies to manage nitrogen loads to estuaries. Because eelgrass restoration may be difficult (Harrison 1990, Davis & Short 1997, Davis et al. 1998), development of indicators of incipient eelgrass decline in response to eutrophication would be useful for directing management efforts towards the prevention of loss. Finally, eelgrass loss and algal overgrowth in estuaries may have important implications for ecosystem production, carbon and nutrient cycling, and trophic linkages to adjacent systems (carbon and nutrient export: Nixon et al. 1986, Cebrián et al. 1998, Cebrián 1999). Understanding the nature and dynamics of the transition from eelgrass to algae under increasing eutrophication is a first step toward understanding the ecological implications associated with that change.

The estuaries of Waquoit Bay, Massachusetts, USA, offer the opportunity to examine the response of estuarine primary producers to increased nitrogen loads at the watershed–estuarine scale. We used the estuaries of Waquoit Bay in a space-for-time substitution (Pickett 1989) to infer the time course of increased eutrophication created by increasing urbanization of watersheds. In the Waquoit Bay estuarine system, different land use patterns within the watersheds of 7 estuaries (Fig. 1), similar in depth and water residence times, have resulted in different annual loads of nitrogen delivered to those estuaries (Table 1). Increased urbanization within certain watersheds is accompanied by increases in delivery of land-derived nitrogen (Table 1). The range of nitrogen loads delivered to Waquoit Bay

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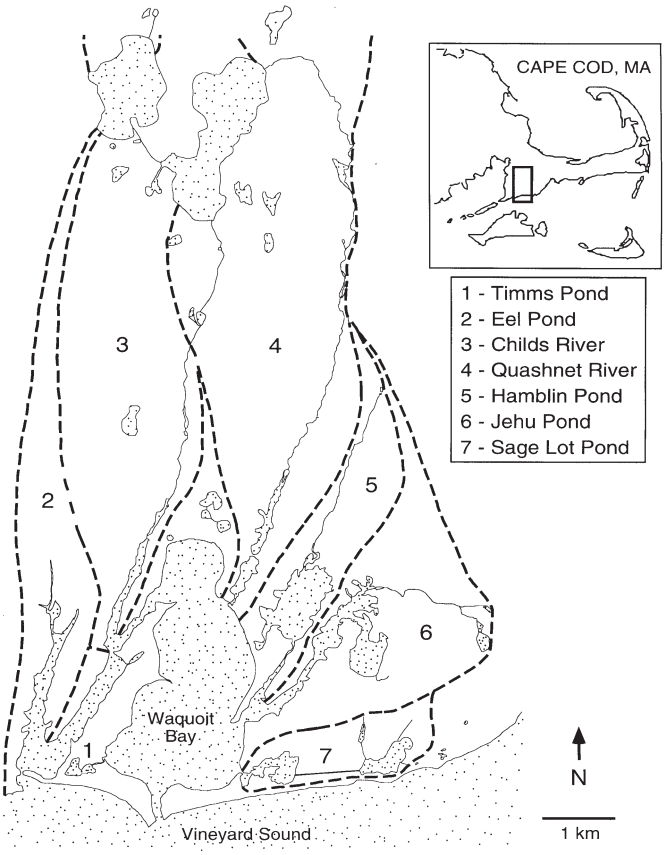

Fig. 1. Map of Waquoit Bay estuarine system (inset: location on Cape Cod, Massachusetts). Watershed delineations for each estu-ary are indicated with dashed lines

estuaries (5 to 407 kg N ha^{-1} yr^{-1}, Valiela et al. 1997a, 2000a) encompasses ~75% of the range of reported loads to different estuaries around the world (Nixon 1992).

In this paper, we make use of inter-estuary comparisons to evaluate the effect of nitrogen supply on (1) the extent of eelgrass bed area and losses of eelgrass habitat over the past decade, and (2) eelgrass mean annual shoot density, biomass and areal biomass, and total annual growth and areal production of leaves, rhizomes, and roots. The effect of nitrogen supply on various aspects of eelgrass productivity is not direct, but seems most likely to be mediated through the stimulation of biomass, and increased light interception by algal producers. To assess this potential indirect effect of nitrogen, we (3) determined annual patterns of phytoplankton, epiphyte, and macroalgal biomass, and (4) estimated how standing stocks of these producers may have modified available irradiance for eelgrass in estuaries subject to different rates of nitrogen loading. Presence of a relationship between these measurements and nitrogen load would yield insight into the mechanisms by which eelgrass decline occurs and reveal potentially useful indicators of incipient decline. We conclude by discussing the implications of our results toward understanding broad-scale ecosystem alterations accompanied by increased nitrogen loading and in the development of management recommendations aimed at preventing eelgrass loss.

MATERIALS AND METHODS

Measurement of eelgrass bed area and recent loss. We determined *Zostera marina* bed area in estuaries subject to different rates of nitrogen loading in September 1997, using a viewbox held over the side of a boat traveling along transects approximately 30 m apart, and running E to W and N to S. Maps in which we delineated eelgrass area in each estuary were scanned and percentage cover was calculated after digitizing the total area of each basin and the coverage of eelgrass (Adobe Photoshop 4.0, Adobe Systems 1996). Loss of eelgrass in the past decade was determined by comparing 1997 maps to those obtained in 1987 by L. Deegan & I. Valiela (pers. obs.) for Sage Lot Pond, and Short & Burdick (1996) for the remainder of the system.

Measurements of eelgrass variables. To evaluate how the seasonal patterns and magnitude of several eelgrass variables may have responded to different

Table 1. Land-derived nitrogen loading rate (normalized for estuarine + salt marsh surface area) (Valiela et al. 1997a, 2000a), number of houses within each watershed (Valiela et al. 1997a, 2000a), mean depth (mean low water + 0.5 m tidal range), and water residence time (Valiela unpubl. data) for the 7 estuaries of Waquoit Bay

Variable	Estuary						
	Timms Pond	Sage Lot Pond	Hamblin Pond	Jehu Pond	Eel Pond	Quashnet River	Childs River
Nitrogen loading rate (kg N ha^{-1} yr^{-1})	5.3	7.6	28.4	30.1	62.7	298	407
Houses ($watershed^{-1}$)	0	0	340	529	718	767	1233
Depth (m)	1.3	1.3	1.5	1.7	1.4	0.8	1.4
Water residence time (d)	1.5	1.5	2.3	2.7	2.0	1.7	2.3

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nitrogen loads, we conducted a field study from November 1997 to November 1998. We routinely measured shoot densities, shoot biomass, areal aboveground biomass, plastochrone intervals, and leaf, rhizome, and root absolute and weight-specific growth rates, and production rates of eelgrass. Measurements were taken every 2 to 8 wk in the 4 estuaries of Waquoit Bay that still supported eelgrass meadows (Timms, Sage Lot, Hamblin Ponds, and Jehu). Of the 7 total estuaries, these 4 received the lowest loads of nitrogen from their watershed. Timms and Sage Lot Ponds have forested watersheds and receive very low loads of land-derived nitrogen (≤8 kg N ha^{-1} yr^{-1}) (Table 1). Jehu and Hamblin Ponds have watersheds that are somewhat urbanized and receive higher loads of nitrogen from their watersheds (~30 kg N ha^{-1} yr^{-1}).

To quantify shoot density, SCUBA divers counted the total number of shoots (vegetative and flowering) within randomly tossed 0.25 m^2 quadrats; 3 to 4 measurements were made in Timms, Sage Lot, and Jehu Ponds, where spatial distributions of shoots were reasonably homogenous, and 6 to 12 measurements were made in Hamblin Pond, where spatial distribution was relatively patchy. We used the marking technique described by Zieman & Wetzel (1980) to measure *in situ* leaf growth rates. To insure that growth rates of all shoot size classes were represented, SCUBA divers tagged all shoots within a given area (at least 25 shoots) and punched 2 holes at the blade–sheath junction of the oldest leaf with a 23-gage hypodermic needle. Shoots were retrieved 2 to 6 wk later (depending on the season), with as much intact rhizome and root material as possible, and a new batch of shoots were tagged and marked. A total of 12 sets of measurements were made throughout the study period. Collected shoots were brought to the laboratory and frozen until processing was possible.

In the laboratory, we ranked the leaves on each shoot by age, and on each leaf we measured total length, width and growth as the distance between the sheath–blade junction and marked holes. We noted whether the tips of the leaves were intact or broken, and used only shoots for which we could unambiguously determine leaf growth (for instance, we discarded shoots where leaf tips were not intact and holes not observed on the remaining leaf blade portion). To convert aboveground shoot characteristics and leaf growth from units of area to biomass, we calculated an average leaf specific density (mg DW cm^{-2} of leaf surface) for each estuary on each date. To determine leaf specific density, we first removed epiphytic material using a glass slide from each leaf on each of 5 shoots, then dried leaves in an oven at 70°C, and weighed them. These values were used to calculate aboveground shoot biomass (mg DW $shoot^{-1}$) and leaf growth rates on a shoot basis (mg DW $shoot^{-1}$ d^{-1}). Weight-specific leaf growth rates were determined by dividing growth rates per shoot by aboveground shoot biomass.

For each marking period, we calculated the plastochrone interval (number of days elapsed between the appearance of 2 consecutive leaves) by dividing the total days elapsed by the mean number of new leaves (i.e. bearing no holes) emerged per shoot (Brouns 1985). Rhizome and root growth rates per shoot (mg DW $shoot^{-1}$ d^{-1}) were calculated from the rate of node formation and growth along the horizontal rhizome, as outlined by Sand-Jensen (1975) and subsequently applied by Pedersen & Borum (1992, 1993) and Duarte et al. (1994). This technique, however, only provides conservative estimates of root growth, since root turnover is fast and the estimates are only based on standing root biomass at the time of collection (Duarte et al. 1998). Shoot-specific rhizome and root growth rates were also determined by dividing growth rates per shoot by aboveground shoot biomass.

Eelgrass aboveground areal biomass was estimated for each sampling interval by multiplying mean shoot density and mean aboveground shoot biomass. We chose not to measure belowground biomass, due to the destructive nature of the collection method, and we only report data for Sage Lot Pond, where long-term above- and belowground biomass data have been taken since 1994 using Eckman grabs (15 cm × 15 cm) (Hersh 1996, Hauxwell et al. 1998, Stieve 2001). Areal leaf, rhizome, and root production were derived by multiplying the corresponding growth rates per shoot by shoot density for each sampling interval, and annual total estimates were derived by summing the production of all compartments over the annual cycle.

Friedman's method for randomized blocks (Sokal & Rohlf 1995) was used to compare shoot densities, shoot biomass, areal aboveground biomass, plastochrone intervals, leaf, rhizome, and root absolute and weight-specific growth rates of eelgrass over time among Timms, Sage Lot, Hamblin, and Jehu Ponds. If significant differences were observed among estuaries, a Wilcoxon's signed-ranks test was employed to determine significant differences between pairs.

Biomass of and light interception by algal producers. To examine the response of algal producers and to quantify their potential effect on light supply for eelgrass, we first determined the biomass of phytoplankton, epiphytes, and macroalgae in each estuary. These data were then used to evaluate potential light interception by each producer. Based on estimates of saturating and compensating irradiances for eelgrass over an annual cycle (a function of temperature), and light availability after attenuation by phytoplankton, epiphytes, and macroalgae, we could compare the re-

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lative importance of potential light limitation of eelgrass in estuaries subject to different rates of nitrogen loading, as detailed below.

Annual measurements of phytoplankton, epiphytes, and macroalgal biomass: During each visit for the eelgrass measurements described above, we also quantified phytoplankton, epiphyte, and macroalgal standing stocks within the 4 estuaries. Measurements of chlorophyll *a* concentrations in the water column were made by collecting replicate 1 l water samples within each eelgrass meadow; in the laboratory, samples were filtered (Whatman GF/F filters), and chlorophyll *a* was determined spectrophotometrically using the method of Lorenzen (1966). Epiphyte biomass was determined for leaves ranked 1 (youngest) to 5, for at least 5 shoots from each estuary. We used a glass slide to scrape epiphytes from each leaf into preweighed aluminum foil envelopes, dried them overnight, and reweighed them. Macroalgal canopy heights were determined using SCUBA and taking 5 to 15 measurements at random within the eelgrass meadows. We used the same statistical approach described for eelgrass annual variables to determine whether phytoplankton, epiphyte, and macroalgal standing stocks differed in estuaries receiving different loads of nitrogen.

Calculations of light attenuation due to phytoplankton, epiphytes and macroalgae: Estimates of light intercepted by each type of producer and resulting irradiance (I) reaching eelgrass surfaces were calculated over the annual cycle as described in Hauxwell et al. (2001). Our general approach was to use measurements of each biological parameter (i.e. phytoplankton, epiphyte, and macroalgal biomass) in equations describing the relationship between biomass and light attenuation for each producer (Bannister 1974, Kirk 1994, Twilley et al. 1985, Peckol & Rivers 1996). Previous studies showed that background attenuation did not vary among different sites in the Waquoit Bay system (Hauxwell et al. 2001), so we used the mean water-column light-attenuation factor for background scatter ($K_B = 0.85\ m^{-1}$) from Hauxwell et al. (2001). Light attenuation due to phytoplankton (K_P) was calculated by multiplying chlorophyll concentrations by a chlorophyll-specific light-attenuation coefficient ($k_c = 0.016\ m^2\ mg^{-1}$ chlorophyll: Bannister 1974, Kirk 1994). Total water-column light-attenuation from surface to depth, z, was calculated according to Beer's law: $I_z/I_0 = e^{-(K_B+K_P)(z)}$. Light attenuation due to mean epiphyte biomass on Leaves 1 to 5 was calculated according to Twilley et al. [1985: Fig. 8, top: $\ln I_z/I_0 = 0.32 - (0.42)$ (epiphyte biomass, in units mg DW cm^{-2} of leaf material)]. Light attenuation due to macroalgal canopies was calculated according to the relationship provided by Peckol & Rivers (1996), in which irradiance decreased exponentially as macroalgal canopy heights increased; by 6 to 8 cm, light penetration was <1% of that reaching the surface of the canopy.

Because eelgrass shoot height has an effect on its depth in the water column and, therefore, the intensity of incoming light it receives, we considered scenarios for average established shoots (>15 cm in height) and also for smaller newly recruiting shoots. For established shoots, we calculated water-column light attenuation as the mean of attenuation between (1) the air-water interface to tips of leaves as shoot height varies throughout a year, and (2) the air-water interface to the sediment-water interface. Epiphyte light attenuation, as described above, was assumed to occur. Macroalgal light attenuation was calculated by multiplying the percentage of photosynthetic material buried by macroalgae (range: 0 to 32% depending on estuary and date, based on mean shoot height and macroalgal canopy height) by light attenuation through the macroalgae canopy. For smaller new shoots, water column light attenuation was calculated from the air-water interface to the sediment-water interface, interception of light by epiphytes was assumed to be negligible, and 100% of the photosynthetic material was assumed to be beneath the macroalgal canopy.

Calculations of epiphytic and macroalgal shading were made assuming that both processes occur simultaneously, and were both based on the magnitude of incoming irradiance after total water-column attenuation. We did not include light interception due to eelgrass canopies, so estimates of resulting light intensities are conservative and only represent the potential modifying effects of water-column, epiphytes, and macroalgae.

Irradiance and prediction of saturating (I_{sat}) and compensating (I_{comp}) irradiances for eelgrass: To convert percentage reductions in irradiance by algal producers to absolute values, we needed to apply irradiance at the air-water interface. Surface irradiance data for the annual cycle were provided by R. Payne of Woods Hole Oceanographic Institution. Based on previous measurements in Waquoit Bay throughout a variety of weather conditions, we assumed a surface reflectance of 35% (Peckol & Rivers 1996).

To assess the effect of light interception by algal producers on eelgrass growth, we compared our estimates of irradiance reaching eelgrass in the Waquoit estuaries to estimates of saturating and compensating irradiance necessary for eelgrass photosynthesis. We determined the annual cycle of I_{sat} and I_{comp} for eelgrass in Waquoit Bay, using the relationships between temperature and I_{sat} and I_{comp} for a nearby Woods Hole eelgrass population (Marsh et al. 1986), applied to Waquoit Bay 1998 temperature data (provided by the Waquoit Bay National Estuarine Research Reserve Baywatchers Program).

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RESULTS

Eelgrass bed area and recent loss in relation to nitrogen loading

For the estuaries that supported eelgrass in 1987 (i.e. those exposed to loading rates between 5 and 63 kg N ha^{-1} yr^{-1}), we regressed eelgrass bed area in 1997 and areal loss between 1987 and 1997 versus nitrogen loading rates (Fig. 2). Across those estuaries, the area of eelgrass habitat decreased logarithmically as nitrogen loading rates increased (Fig. 2, top). Similarly, loss of eelgrass habitat, expressed as a percentage of the existing area in 1987, increased logarithmically with higher loads (Fig. 2, bottom).

Loss of eelgrass habitat from estuaries of Waquoit Bay between 1987 and 1997 was extensive. Eelgrass disappeared entirely from Eel Pond, the estuary with the highest nitrogen loading rate (63 kg N ha^{-1} yr^{-1}) that still supported eelgrass in 1987. In Hamblin and Jehu Ponds, where nitrogen loading rates were ~30 kg N ha^{-1} yr^{-1}, eelgrass nearly disappeared. Only ~6500 of the ~180 000 m^2 of eelgrass present in 1987 remained in Hamblin Pond, a loss of >96%. In Jehu Pond, 85% of the area in 1987 was lost, with only ~20 000 m^2 remaining out of the ~130 000 m^2 of eelgrass habitat present in 1987. There was some loss of eelgrass habitat (13 to 32%) even in Timms Pond and Sage Lot Pond, where nitrogen loads were low (≤8 kg N ha^{-1} yr^{-1}). In 1997, Timms Pond and Sage Lot supported 14 000 and 30 000 m^2 of eelgrass habitat, respectively.

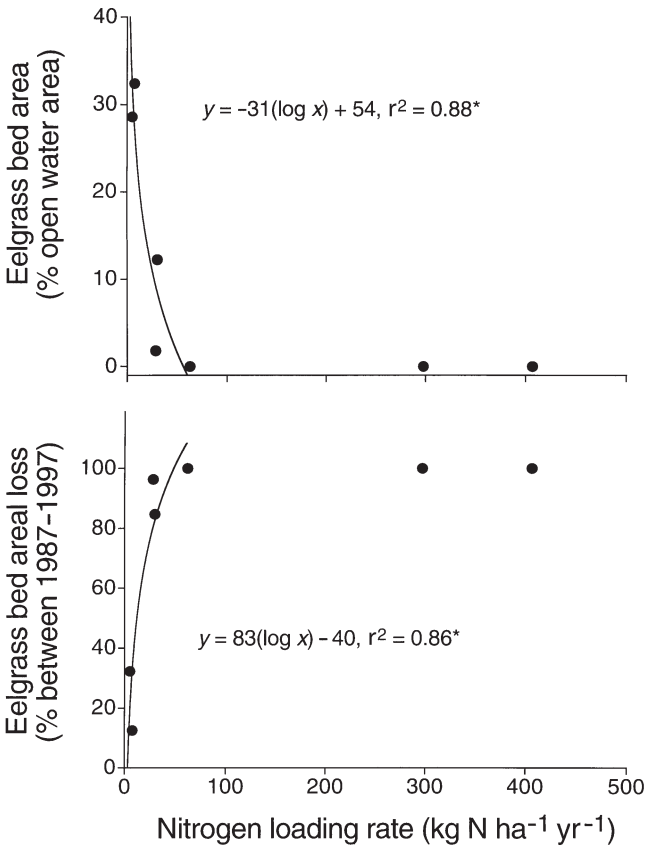

Fig. 2. *Zostera marina.* Eelgrass areal cover (top) and areal loss (bottom) between 1987 and 1997 in estuaries of the Waquoit Bay system subject to different rates of nitrogen loading. Regression analyses were conducted within the nitrogen loads represented by the estuaries that still had eelgrass in 1987 (Timms, Sage Lot, Hamblin, Jehu, and Eel Ponds: 5 to 63 kg N ha^{-1} yr^{-1}). Here and in later figures, * $p < 0.05$

Eelgrass annual variables in relation to nitrogen loading

Annual means of our eelgrass measurements are summarized in Table 2. Of all the eelgrass measurements, shoot density, shoot and areal aboveground biomass, leaf growth rate, and leaf, rhizome, and root production differed among the 4 estuaries (Table 3). For all of these measurements, the meadow in Sage Lot Pond (a low-nitrogen site) consistently represented the highest values recorded, while values for the Hamblin Pond meadow (a higher-nitrogen site) were the lowest (Table 2). Values for Timms Pond (a low-nitrogen site) and Jehu Pond (a higher-nitrogen site), however, were aligned with the other estuary of similar nitrogen load (Sage Lot Pond or Hamblin Pond, respectively) only in the cases of shoot density, aboveground areal biomass, and production by leaves, rhizomes, and roots (Table 2). Within-meadow differences in shoot density contributed to the differences observed in areal biomass and production rates within meadows, which were arithmetically derived by multiplying shoot density by shoot biomass or shoot growth rates, variables that were not aligned in relation to nitrogen load (Table 3).

Regression analyses were conducted to ascertain which eelgrass variables might be indicators of decline associated with increased nitrogen loading before disappearance of habitat. These regressions were, therefore, restricted to the narrow, relatively low range of loads represented by estuaries that supported eelgrass within the past decade (0 to 63 kg N ha^{-1} yr^{-1}). Mean annual shoot density and aboveground areal biomass (within meadows) decreased exponentially as nitrogen loading rates increased (Fig. 3). Total annual eelgrass production within meadows decreased exponentially as nitrogen loading rates increased (Fig. 4, top). Total eelgrass production within the estuary, calculated as the sum of the products between within-meadow leaf, rhizome, and root production applied to the area of eelgrass bed in the estuary, also decreased exponentially as nitrogen loading rates increased (Fig. 4, bottom).

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Table 2. *Zostera marina*. Summary of results of annual measurements of shoot densities (annual within-meadow mean ± SE), aboveground shoot biomass (annual mean ± SE), aboveground areal biomass (annual within-meadow mean ± SE), number of plastochrone units produced (annual total), and leaf, rhizome and root absolute and weight-specific growth rates (annual total ± SE) and production rates (annual within-meadow total ± SE) of eelgrass in 4 estuaries of Waquoit Bay subject to different rates of nitrogen loading

Variable	Estuary			
	Timms Pond	Sage Lot Pond	Hamblin Pond	Jehu Pond
Density (shoots m^{-2})	196 ± 25	266 ± 28	57 ± 5	141 ± 23
Aboveground shoot biomass (mg DW $shoot^{-1}$)	130 ± 24	253 ± 47	160 ± 19	244 ± 41
Aboveground areal biomass (g DW m^{-2})	32 ± 8	80 ± 19	9 ± 1	36 ± 9
Plastochrone units (number yr^{-1})	30.4	27.2	31.1	29.9
Leaf growth rate (mg DW $shoot^{-1}$ yr^{-1})	690 ± 30	1280 ± 80	870 ± 60	1169 ± 60
Rhizome growth rate (mg DW $shoot^{-1}$ yr^{-1})	210 ± 10	240 ± 20	220 ± 10	270 ± 10
Root growth rate (mg DW $shoot^{-1}$ yr^{-1})	110 ± 5	170 ± 10	180 ± 10	180 ± 20
Weight-specific leaf growth rate (% yr^{-1})	574 ± 13	523 ± 10	562 ± 17	493 ± 12
Weight-specific rhizome growth rate (% yr^{-1})	200 ± 15	122 ± 9	150 ± 23	117 ± 24
Weight-specific root growth rate (% yr^{-1})	142 ± 10	111 ± 7	129 ± 14	94 ± 16
Leaf production (mg DW m^{-2})	159 ± 9	395 ± 29	49 ± 6	203 ± 7
Rhizome production (mg DW m^{-2})	44 ± 3	74 ± 6	13 ± 2	54 ± 3
Root production (mg DW m^{-2})	20 ± 1	46 ± 3	12 ± 1	30 ± 3

Table 3. *Zostera marina*. Results of Friedman's method for randomized blocks (Statview®, SAS Institute 1999) used to compare shoot densities, aboveground shoot biomass, aboveground areal biomass, plastochrone intervals, and leaf, rhizome and root absolute and weight-specific growth rates and production rates of eelgrass over time in 4 estuaries of Waquoit Bay subject to different rates of nitrogen loading (ns: not significant, $p > 0.05$; $^{**}p < 0.01$; $^{***}p < 0.001$). When significant differences were observed among sites, a Wilcoxon's signed-ranks test (Statview®) was employed to compare pairs. Results of the ranking were presented in order from highest to lowest; =: no significant difference between adjacent pairs; >: significant differences ($p < 0.05$) between adjacent pairs (S: Sage Lot Pond, T: Timms Pond, J: Jehu Pond, H: Hamblin Pond)

Variable	p	Statistical ranking
Density (shoots m^{-2})	***	S > T = J > H
Aboveground shoot biomass (mg DW $shoot^{-1}$)	**	S = J > T = H
Aboveground areal biomass (g DW m^{-2})	***	S > J = T > H
Plastochrone interval (d $leaf^{-1}$)	ns	
Leaf growth rate (mg DW $shoot^{-1}$ d^{-1})	**	S = J = H = T, S > H, S > T, J > T
Rhizome growth rate (mg DW $shoot^{-1}$ d^{-1})	ns	
Root growth rate (mg DW $shoot^{-1}$ d^{-1})	ns	
Weight-specific leaf growth rate (% d^{-1})	ns	
Weight-specific rhizome growth rate (% d^{-1})	ns	
Weight-specific root growth rate (% d^{-1})	ns	
Leaf production (mg DW m^{-2})	***	S > J = T>H
Rhizome production (mg DW m^{-2})	***	S > J = T>H
Root production (mg DW m^{-2})	**	S = J = T > H, S > T

Light interception by algal producers and relationship to nitrogen load

Annual measurements of phytoplankton, epiphytes and macroalgal biomass

Phytoplankton biomass peaked at the end of August in all estuaries between 11 and 23 µg chlorophyll *a* l^{-1}, and was ≤4 mg chlorophyll *a* l^{-1} from November to May (Fig. 5, top). Epiphyte biomass was lowest in the spring when plastochrone intervals were at a minimum, and accumulation of epiphytic material was reduced due to the relatively fast appearance of new leaves and shedding of old leaves (Fig. 5, middle). Epiphyte biomass was highest during late summer, fall, and early winter when plastochrone intervals were longer and hence, the time in which epiphytes might colonize leaves was longer (Fig. 5, middle). The highest peak in epiphyte biomass was in Hamblin Pond in late August at 13 mg DW cm^{-2} of leaf material. Macroalgal biomass fluctuated over the year in all estuaries and was generally highest in Hamblin Pond (4 to 10 cm) and lowest in Timms Pond (0 to 2 cm) (Fig. 5, bottom). Macroalgal blooms occurred in Jehu Pond during December 1997 and in November 1998 when mean canopy heights reached 13 cm.

Of these measurements, macroalgal canopy height was the only variable to differ significantly among estuaries (Table 4). Maximum macroalgal canopy height increased linearly as nitrogen loading rate increased, within the range of loads in which eelgrass was still present, and also across the entire range of loads present to the Waquoit Bay estuaries (Fig. 6). Canopies were primarily comprised of the fast-growing, nutrient-limited taxa *Cladophora vagabunda* and *Gracilaria tikvahiae* (Peckol et al. 1994).

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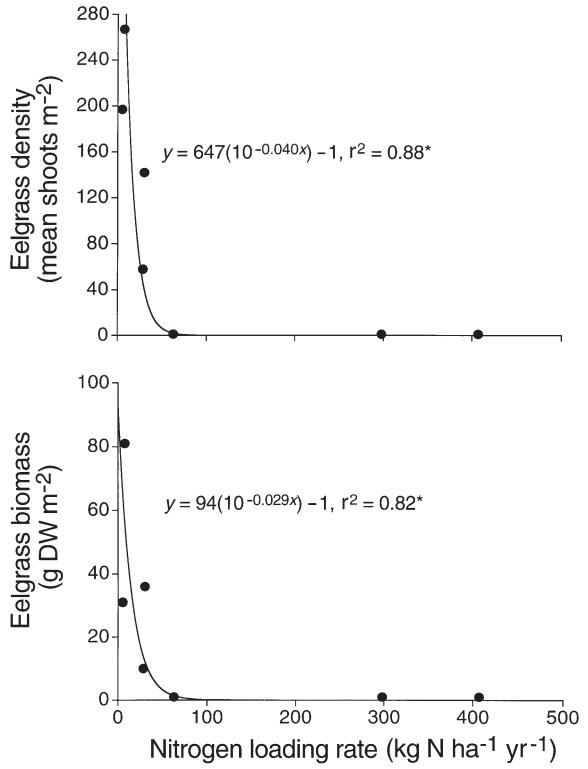

Fig. 3. *Zostera marina.* Mean annual 1998 within-meadow shoot density (top) and areal biomass (bottom) of eelgrass in estuaries of the Waquoit Bay system subject to different rates of nitrogen loading. Regression analyses were conducted within the nitrogen loads represented by the estuaries that still had eelgrass in 1987 (Timms, Sage Lot, Hamblin, Jehu, and Eel Ponds: 5 to 63 kg N ha^{-1} yr^{-1})

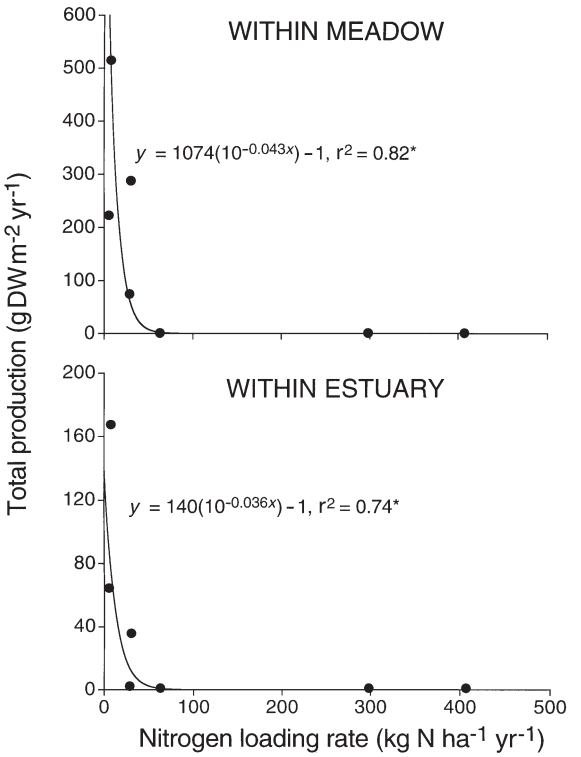

Fig. 4. *Zostera marina.* Total annual 1998 eelgrass areal production within meadows and normalized to bed area within estuaries of the Waquoit Bay system subject to different rates of nitrogen loading. Regression analyses were conducted within the nitrogen loads represented by the estuaries that still had eelgrass in 1987 (Timms, Sage Lot, Hamblin, Jehu, and Eel Ponds; 5 to 63 kg N ha^{-1} yr^{-1})

Light attenuation due to phytoplankton, epiphytes and macroalgae

Estimated light attenuation through the water column from the surface to the sediment-water interface remained relatively constant throughout the year, ranging from 68 to 82% (Fig. 7, top right). The small peak at the end of August was due to the high-standing stocks of phytoplankton in all estuaries at that time (Fig. 5, top). Due to the increase in eelgrass shoot height from spring to summer (shoots ranged from 15 to 95 cm in height over the year) and hence, shallower depth (z), estimated water-column light attenuation from the surface to leaf tips was lowest during late spring and early summer (Fig. 7, top left).

Table 4. Results of Friedman's method for randomized blocks (Statview®, SAS Institute 1999) used to compare standing stocks of phytoplankton, epiphytes, macroalgae, and estimated irradiance reaching established and new eelgrass *Zostera marina* shoots over time in 4 estuaries of Waquoit Bay subject to different rates of nitrogen loading (ns: not significant, $p > 0.05$; ***$p < 0.001$). If significant differences were observed among sites, a Wilcoxon's signed-ranks test (Statview®) was employed to compare pairs. Results of the ranking were presented in order from highest to lowest; =: no significant difference between adjacent pairs; >: significant differences ($p < 0.05$) between adjacent pairs (S: Sage Lot Pond, T: Timms Pond, J: Jehu Pond, H: Hamblin Pond)

Variable	p	Statistical ranking
Phytoplankton (µg chlorophyll a l^{-1})	ns	
Epiphyte biomass (mg cm^{-2})	ns	
Macroalgal canopy height (cm)	***	H > J = S > T
Irradiance reaching established eelgrass shoots (µmol photons m^{-2} s^{-1})	***	S > T = J = H, T > H
Irradiance reaching new eelgrass shoots (µmol photons m^{-2} s^{-1})	***	T > S = J > H

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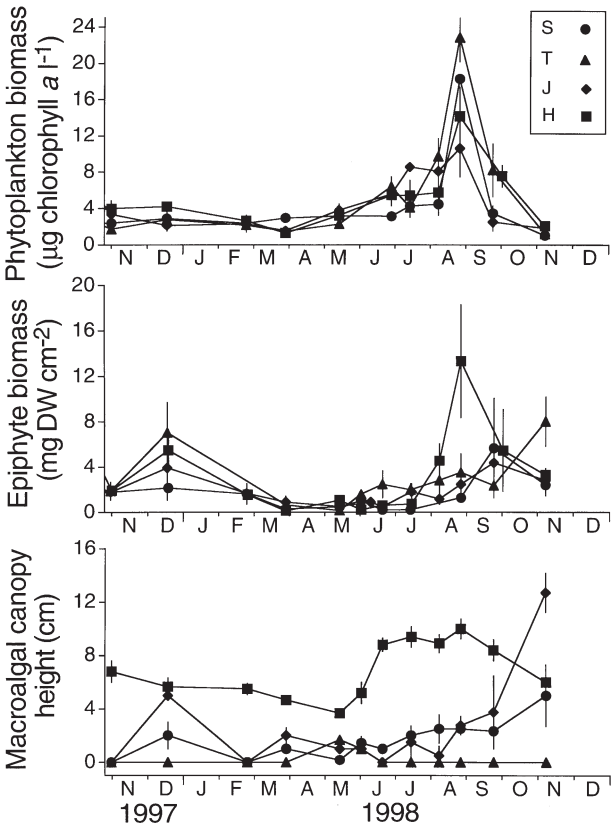

Fig. 5. Standing stocks of phytoplankton (top), epiphytes of eelgrass *Zostera marina* (middle), and macroalgae (bottom) between November 1997 and November 1998 in 4 estuaries of Waquoit Bay subject to different rates of nitrogen loading (S: Sage Lot Pond, T: Timms Pond, J: Jehu Pond, H: Hamblin Pond)

Estimated light attenuation by epiphytes for established shoots ranged from 0 to 100% (Fig. 7, middle left) after accounting for water-column attenuation. During peaks in epiphyte biomass, light attenuation by epiphytes may have been of greater importance than water column attenuation for established shoots (Fig. 7, top left). Since epiphyte biomass was negligible for new shoots, light attenuation was 0% (Fig. 7, middle right).

Estimated light attenuation by macroalgal canopies was relatively minor for established shoots, ranging from 0 to 31% (Fig. 7, bottom left) after accounting for water-column attenuation. For new shoots, however, light attenuation by macroalgal canopies may have been severe; the lowest peak occurred in Timms Pond at 77% (Fig. 7, bottom right). Estimated light attenuation in Hamblin Pond was 91 to 100% throughout the annual cycle (Fig. 7, bottom right).

After attenuation by the water column, epiphytes, and macroalgae, estimated irradiance available for photosynthesis by established eelgrass shoots was highest for the Sage Lot Pond meadow and lowest for the Hamblin Pond meadow (Fig. 8, middle). Our conservative estimates yielded irradiance above saturating levels for established shoots in all meadows between January and August. During late summer and fall, established shoots may have received between compensating and saturating levels of irradiance, except in Hamblin Pond where established shoots may have received less than compensating levels of irradiance.

Estimated irradiance available for photosynthesis by new shoots was highest (above or near saturation) in Timms Pond, where macroalgal canopies were lowest (Fig. 8, bottom). Estimates for Sage Lot and Jehu Ponds ranged between compensating and above-saturating levels of irradiance. Estimates were lowest in Hamblin Pond, at or below compensating levels of irradiance for all but 1 date in the annual cycle.

Over an annual cycle, the estimated irradiance which reached established and new shoots varied significantly among estuaries (Table 4). The mean annual irradiance which reached established and new eelgrass shoots was significantly lower in estuaries of higher-nitrogen loads (Fig. 9). Within estuaries of similar nitrogen load, established shoots were estimated to receive significantly higher irradiance than new shoots (paired *t*-tests for values throughout the annual cycle, $p < 0.05$).

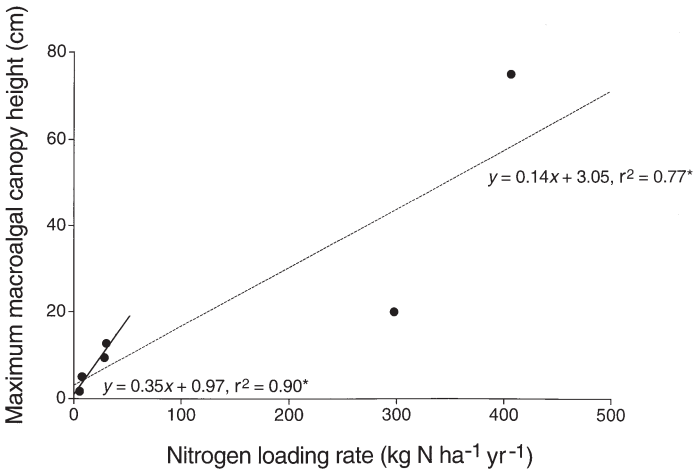

Fig. 6. Maximum canopy height of macroalgae during 1998 in estuaries of the Waquoit Bay system subject to different rates of nitrogen loading. Regression analyses were conducted for the 4 estuaries in which we took measurements (Timms, Sage Lot, Hamblin, and Jehu Ponds: 5 to 30 kg N ha^{-1} yr^{-1}) and for the entire range of values present in the Waquoit Bay system (dashed line) including Childs and Quashnet Rivers (5 to 407 kg N ha^{-1} yr^{-1}; data from Hauxwell et al. (2001)

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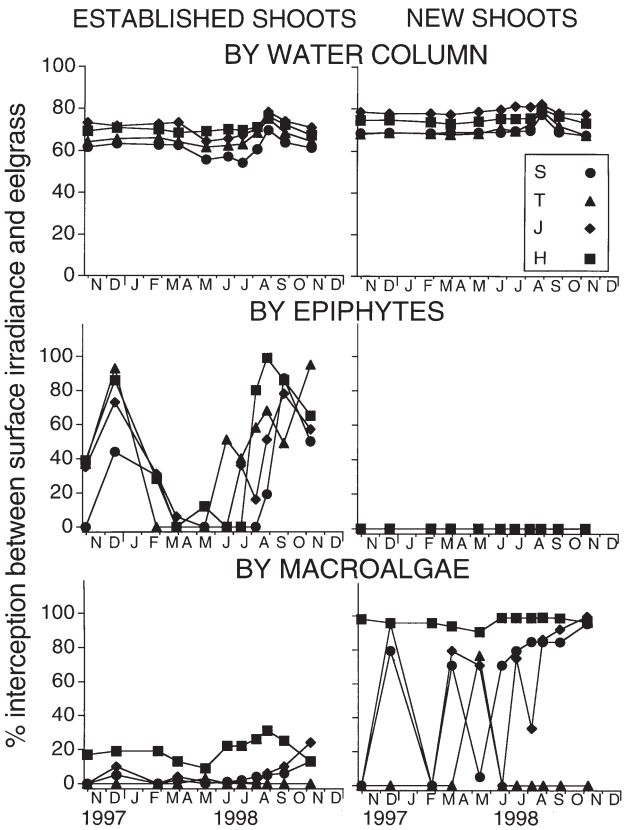

Fig. 7. *Zostera marina.* Percentage of surface irradiance attenuated through the water column (top), by epiphytes (middle), and by macroalgal canopies (bottom) for established (left) and newly recruiting (right) eelgrass shoots between November 1997 and November 1998 in 4 estuaries of Waquoit Bay subject to different rates of nitrogen loading (S: Sage Lot Pond, T: Timms Pond, J: Jehu Pond, H: Hamblin Pond). Epiphyte and macroalgal light attenuation were based on incoming irradiance after water-column attenuation

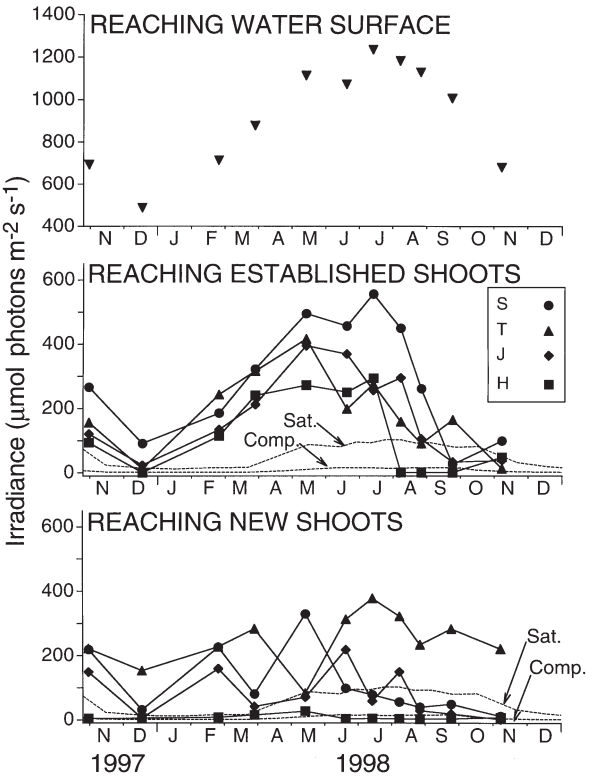

Fig. 8. *Zostera marina.* Monthly surface irradiance (mean for daylight hours) corrected for reflectance (top) and estimates of available irradiance (after attenuation by the water column, epiphytes, and macroalgal canopies) to established (middle) or new (bottom) eelgrass shoots between November 1997 and November 1998 in 4 estuaries of Waquoit Bay subject to different rates of nitrogen loading (S: Sage Lot Pond, T: Timms Pond, J: Jehu Pond, H: Hamblin Pond) (surface irradiance data from R. Payne, Woods Hole Oceanographic Institution). Saturating (Sat.) and compensating (Comp.) levels of irradiance for eelgrass, corresponding to actual temperatures over the annual cycle are also shown

DISCUSSION

Nitrogen loading and eelgrass loss

Nitrogen loading to the Waquoit Bay system has contributed to extensive loss of eelgrass *Zostera marina* (Figs. 2–4). Results of this large-scale comparison at the watershed–estuarine scale are in agreement with previous laboratory and mesocosm experiments (Short et al. 1993,1995), i.e. they both show that (1) eelgrass is very sensitive to eutrophication, with large losses occurring rapidly even at relatively low-nitrogen loading rates, and (2) light limitation imposed by nutrient-limited algae is an important mechanism by which such losses may occur. Substantial eelgrass loss was observed in all Waquoit Bay estuaries, except in those with the lowest loading rates (Timms and Sage Lot Ponds; Fig. 2). In the Waquoit estuaries, nitrogen loads that allow eelgrass survival appear to be <28 to 63 kg N ha^{-1} yr^{-1}. This load matches hindcast estimates of nitrogen loading (based on historical land-use data and the Waquoit Bay Nitrogen Loading Model, Valiela et al. 2000a) to now eutrophic subestuaries for the period of time when eelgrass loss was documented historically (Bowen & Valiela 2001). Presence of seed coats in cores taken from estuaries with the highest loads of nitrogen revealed that Childs River and Quashnet River, estuaries now receiving loads between 298 and 407 kg N ha^{-1} yr^{-1}, once supported eelgrass prior to extensive residential development of their watersheds (Safran et al. 1998). Seagrasses in general appear to be sensitive indicators of nitrogen loading. In a compilation by Valiela & Cole (2002) of worldwide seagrass loss over a range of reported nitrogen loads, an identical general

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pattern was observed, with substantial losses (>50%) occurring within 50 to 100 kg N ha^{-1} yr^{-1} and total disappearance at loadings exceeding 100 kg N ha^{-1} yr^{-1}.

In Waquoit Bay, loss of eelgrass under increasing nitrogen loads seems to occur mainly through a decrease in shoot density (Fig. 3, top), rather than as a result of reduced growth rates per shoot (Tables 2 & 3). Under higher-nitrogen loads (Hamblin and Jehu Ponds), we observed rapid loss of eelgrass bed area (Fig. 2) and relatively low shoot densities (Table 2, Fig. 3, top); however, we found no significant relationship between nitrogen loading rates and the above- or belowground growth rates of established shoots (Table 2). Hence, across the estuaries compared, more eutrophic estuaries have fewer shoots per unit of meadow area, but the remaining shoots generally grow at the same rate as those in low-nitrogen estuaries. These differences suggest reduced shoot recruitment or promoted shoot mortality as plausible mechanisms leading to eelgrass decline in eutrophic estuaries. This discrepancy between density data and growth data implies that established shoots function similarly on a per shoot basis among estuaries, but that established shoots from higher-nitrogen estuaries lack sufficient resources to translocate energy to newly recruiting clonal branches. Hence, loss occurs at both the edges of a meadow and within a meadow.

The speculation that diminished recruitment accounts for eelgrass decline is further supported by our estimates of light availability to new shoots (Figs. 8, bottom & 9). From light budgets which considered water column, epiphyte, and macroalgal shading, we estimated chronic, severe light limitation to newly recruiting shoots in Hamblin Pond, due mainly to shading by a coexisting 4 to 10 cm macroalgal canopy (Figs. 5, bottom & 8, bottom). Recruiting shoots were exposed to under-compensating irradiances for most of the year in Hamblin Pond due to a high, persistent macroalgal canopy, and during the 1998 fall in Jehu Pond due to a concurrent macroalgal bloom (Fig. 8). Severe light limitation of recruiting shoots, however, was not observed in the 2 low-nitrogen estuaries. Additional deleterious effects associated with large macroalgal canopies may occur via unfavorable biogeochemical conditions imposed on buried eelgrass shoots, such as anoxia (Pregnall et al. 1984, Koch et al. 1990), other redox changes resulting from low-oxygen concentration (i.e. high sulfide concentrations: Goodman et al. 1995, Terrados et al. 1999), and toxic ammonium concentrations (Van Katwijk et al. 1997). In fact, the capacity for large macroalgal canopies (>12 cm) to preclude eelgrass shoot recruitment has been experimentally demonstrated by Hauxwell et al. (2001). Our results also contribute evidence for increased mortality of established shoots in higher-nitrogen estuaries due to intense shading by epiphytes; established shoots in Hamblin Pond were estimated to receive less than compensating irradiance during the fall of the two years of survey (i.e. highest epiphyte biomass). Nitrate concentrations in these estuaries were below toxic levels (Burkholder et al. 1992,1994, Hauxwell et al. 2001)

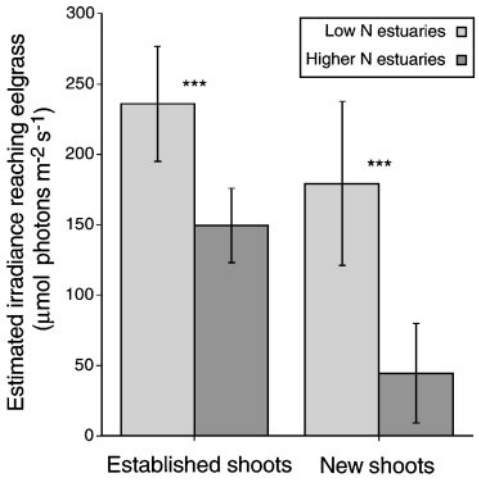

Fig. 9. *Zostera marina.* Estimated mean (±SE) annual 1998 irradiance that reached established and new shoots of eelgrass in estuaries of Waquoit Bay receiving low (5 to 8 kg N ha^{-1} yr^{-1}, Timms and Sage Lot Ponds as replicates) versus higher (28 to 30 kg N ha^{-1} yr^{-1}, Hamblin and Jehu Ponds as replicates) loads of land-derived nitrogen. Paired *t*-tests for values throughout the annual cycle were used to determine whether there were significant differences between low and higher-nitrogen estuaries (***$p < 0.001$)

Coastal management

Management of eelgrass habitats is often mandated by state and local governments. In Massachusetts, for instance, state regulations dictate that stakeholders in projects involving dredging, filling, or altering parcels of coastland must first demonstrate that they will minimize deleterious impacts, or have no adverse effects on eelgrass beds. In addition, there are other stringent local bylaws on land development, such as regulations that often make dock construction illegal. The very important link, however, between watershed development >30 m from the water's edge and adjoining estuarine eelgrass health/water quality is largely unregulated. Watershed influences on nitrogen load arguably have more far-reaching negative impacts on eelgrass habitat and water quality (Figs. 2–6), and these issues continue to be addressed on a regional/local level on Cape Cod.

Two general recommendations emerge from this work for managers investing in eelgrass preservation. First, since this comparison and others (Valiela et al. 2000b,

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Valiela & Cole 2002) show that eelgrass is lost within a relatively low and narrow range of nitrogen loading rates, watersheds should be developed or managed such that land-derived loads are kept low. The threshold value necessary for eelgrass preservation is difficult to establish accurately, since many factors may influence land-derived nitrogen loading and fate in estuaries (i.e. retention by surrounding marsh, water residence time: Valiela et al. 2000a, 2001), but the present results and others (Valiela et al. 2000b, Valiela & Cole 2002) suggest that eelgrass is likely to decline substantially at values <30 to 100 kg N ha^{-1} yr^{-1}. Several strategies, some economically feasible and some not, within already-developed communities may reduce nitrogen loads to such levels (Valiela et al. 2000b: sewering towns, green space/salt marsh preservation, minimizing fertilizer use, etc.).

Our second recommendation involves assessing eelgrass health. Because eelgrass restoration is difficult (Harrison 1990, Davis & Short 1997, Davis et al. 1998), simple but accurate indicators of incipient eelgrass decline due to nitrogen loading are needed. Since depressed shoot recruitment and increased mortality seem to be important processes in eelgrass decline, we recommend routine monitoring of shoot density within meadows and, if possible, eelgrass bed area. Since algae, like eelgrass, are very sensitive indicators of nitrogen loading (Fig. 6), routine monitoring of macroalgal distributions/canopy heights and Secchi depths (in estuaries with longer residence times) may also prove useful.

In contrast, shoot growth measurements require SCUBA, are time-consuming, and did not yield a relationship with nitrogen loading rate in the meadows we studied. Morphological features of eelgrass and shoot biomass vary widely among and within stable populations (van Lent & Verschuure 1994) and may not be useful indicators. Physiological measurements of eelgrass tissues (C:N, [chlorophyll *a*]) require access to expensive scientific equipment and again, do not necessarily yield consistent comparative information (J. Hauxwell et al. unpubl. data), although C:N ratios have been hypothesized to be potential indicators of nutrient availability (Fourqurean et al. 1997). Recent findings by McClelland & Valiela (1998) suggested that stable isotopic nitrogen signatures ($\delta^{15}N$) measured in estuarine primary producers may be useful in detecting wastewater-derived nitrogen loading. Over a broad range of nitrogen loading rates (5 to 407 kg N ha^{-1} yr^{-1}), this signature could even be used to estimate wastewater-nitrogen loading rates. Preliminary evidence, however, suggests that this approach is not sensitive enough to detect differences in wastewater input within the low and narrow range of nitrogen loads in which eelgrass disappears. Stable isotopic nitrogen signatures measured in macroalgae from Sage Lot and Hamblin Ponds do not consistently reflect the slight but crucial increase in wastewater nitrogen inputs that apparently contribute to eelgrass decline (Table 5). A more thorough investigation of this approach in detecting wastewater nitrogen inputs across the relative small loading range relevant to eelgrass decline is highly recommended and may still reveal this technique to be useful for the management of eelgrass preservation.

Relative contribution by eelgrass, macroalgae and phytoplankton to total primary productivity under low- or high-nitrogen loads

Our comparison shows a shift from eelgrass-dominated to macroalgal-dominated communities following increased eutrophication, similar to the results of past field comparisons and manipulative experiments (Kemp et al. 1983, Twilley et al. 1985, Sand-Jensen & Borum 1991, Duarte 1995, Short et al. 1995). To examine the implications of such shift on total primary production in Waquoit Bay estuaries, we compared annual eelgrass, macroalgae, and phytoplankton production, standardized to estuarine area, in 2 estuaries representing extremes of the nitrogen loading gradient found in Waquoit Bay (Table 6). Under low-nitrogen conditions, eelgrass production within the meadow (i.e. scaled to m^2 of meadow, 515 g m^{-2} yr^{-1}) was similar in magnitude to that of macroalgae and approximately twice that of phytoplankton. However, after extrapolating to m^{-2} of estuarine area, production by eelgrass was lower than that by both macroalgae and phytoplankton, even under low-nirogen conditions. Under high-nitrogen conditions, eelgrass disappeared, macroalgal production almost tripled, and phytoplankton production more than doubled.

In the Waquoit system, total primary production in the estuary exposed to the highest annual load of nitrogen was more than twice that in the estuary exposed to the lowest load of nitrogen (Table 6). Replacement of eelgrass habitat by macroalgal- and phytoplankton-

Table 5. $\delta^{15}N$ values in macroalgae from 2 estuaries of Waquoit Bay subject to different rates of nitrogen loading. Macroalgal samples were composites of specimens collected from 5 sites within each estuary in November 1999 (means ± SE of replicate composites)

Species	Estuary	
	Sage Lot Pond	Hamblin Pond
Cladophora vagabunda	3.5 ± 0.09	4.1 ± 0.09
Gracilaria tikvahiae	5.8 ± 0.05	5.6 ± 0.06
Codium fragile	6.1 ± 0.07	6.5 ± 0.07
Fucus vesiculosis	4.8 ± 0.00	4.2 ± 0.07

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Table 6. Comparison of annual net production estimates of eelgrass *Zostera marina* (normalized to estuarine surface area), macroalgae (Hauxwell et al. [1998], extended for the annual cycle), and phytoplankton (J. H. Foreman et al. unpubl. data) in 2 estuaries of Waquoit Bay subject to relatively low (Sage Lot Pond, 0.5 g N m^{-2} yr^{-1}) or high (Childs River, 36 g N m^{-2} yr^{-1}) rates of land-derived nitrogen loading (see Table 1). Production is expressed in terms of biomass, carbon fixation, and nitrogen incorporation (g DW m^{-2} yr^{-1}). To convert from biomass units to carbon fixation or nitrogen incorporation, we assumed carbon and nitrogen content relative to eelgrass biomass to be 40 and 3%, respectively (J. Hauxwell pers. obs.). Carbon and nitrogen content relative to macroalgal biomass were 25 and 2–3%, respectively (Hauxwell et al. 1998). For phytoplankton, we assumed a carbon to biomass ratio of 0.4 and a C:N molar ratio of 7:1 (Redfield 1958)

Producer	Biomass		Carbon		Nitrogen	
	low-nitrogen	high-nitrogen	low-nitrogen	high-nitrogen	low-nitrogen	high-nitrogen
Eelgrass	167	0	67	0	5	0
Macroalgae	725	2071	180	518	22	62
Phytoplankton	235	583	94	233	15	39
Total	1127	2654	341	751	42	101

dominated communities under high rates of nitrogen loading may have resulted in a 120% increase in total carbon fixation. Annual measurements of net ecosystem production for these estuaries corroborate this difference; ecosystem net production was 4-fold greater in the high-nitrogen versus the low-nitrogen estuary (D'Avanzo et al. 1996). These results are contrary to the non-relationship between total primary production and nitrogen loading rates recorded by Borum & Sand-Jensen (1996), who found that for many systems phytoplankton productivity was stimulated, but that benthic production declined as nitrogen loading rates increased. This was probably due to phytoplankton shading of macroalgae, rendering them less productive than the seagrass systems they replace. Two factors may explain the discrepancy between our results and the findings of Borum & Sand-Jensen: (1) for the shallow Waquoit system, macroalgae have higher areal rates of production in estuaries of higher-nitrogen loads despite stimulated phytoplankton growth; (2) eelgrass production in the Waquoit system under low-nitrogen loads is relatively low (515 g DW m^{-2} of meadow yr^{-1}) compared to the worldwide average (J. Hauxwell et al. unpubl. data: 1145 g DW m^{-2} yr^{-1}) due to low shoot densities relative to other populations. Overall, these shifts in quantity and quality of organic material imply ecologically significant consequences in terms of carbon fixation and the various fates of production (changes in rates of herbivory, decomposition, storage, and export).

As in several other systems for which nitrogen budgets have been estimated (summarized in Nixon & Pilson 1983), estimated nitrogen incorporation by all producers in the Waquoit estuaries exceeded land-derived inputs (Table 6). Since eelgrass may be efficient at reclaiming nitrogen from senescent leaf material (Borum et al. 1989, Pedersen & Borum 1992), our estimate of nitrogen incorporation for eelgrass may be high (Table 6). Pedersen & Borum (1993) estimated total nitrogen uptake to be 73% from external sources and 27% from internal recycling. Hence, a more realistic estimate of eelgrass nitrogen demand in the low-nitrogen estuary might be approximately 3.7 g N m^{-2} yr^{-1}. Overall, total nitrogen demand for primary production shifted from ~42 to 101 g N m^{-2} yr^{-1} across the range of nitrogen loads encountered in Waquoit Bay. Since land-derived loading rates of nitrogen were only 0.5 g N m^{-2} yr^{-1} in the low-nitrogen estuary and 36 g N m^{-2} yr^{-1} in the high-nitrogen estuary, regenerated nitrogen and/or nitrogen imported in seawater during tidal exchange must have supported 99% of nitrogen production in the low-nitrogen estuary and 64% in the high-nitrogen estuary. Of the 13 estuaries summarized in Nixon & Pilson (1983), only the lower New York Bay and a section of San Francisco Bay received more 'new' nitrogen from land than was required by primary producers; for the remaining estuaries, nitrogen regeneration was estimated to support 50 to 91% of primary production, similar to the range represented by the Waquoit system.

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