FINAL REPORT TO THE NATURE CONSERVANCY, JUNE 2009

AN ASSESSMENT OF THE IMPACT OF NUTRIENT LOADING, BIVALVE FILTRATION AND PHYTOPLANKTON COMMUNITIES ON ESTUARINE RESOURCES

Christopher J. Gobler, Bradley J. Peterson, Charles C. Wall

Stony Brook University, School of Marine and Atmospheric Sciences

Contents:

Chapter I:

The effects of eutrophication within Peconic Estuary on the growth and survival of estuarine shellfish and seagrass (*Zostera marina*, *Mercenaria mercenaria*, *Crassostrea virginica*, *Argopecten irradians*)

This was a field study conducted during the summer of 2008 and will be submitted as a manuscript to an international peer-reviewed journal this year.

Keywords: *Zostera marina, Crassostrea virginica, Mercenaria mercenaria, Argopecten irradians, Crepidula fornicata, Cyprinodon variegatus,* eelgrass, seagrass, clams, oysters, scallops eutrophication, nutrients, nutrient-loading, bivalves, suspension feeders, ecosystem-based management, estuarine restoration, Peconic Bay, Peconic Estuary

Abstract: While many coastal ecosystems previously supported dense meadows of seagrass and dense stocks of resource bivalves, the impacts of overfishing, eutrophication, harmful algal blooms, and habitat loss have contributed to the decline of these important resources. Anthropogenic nutrient loading that leads to eutrophication has been identified by some researchers as a primary driver of these losses, but other researchers have described potential positive effects of eutrophication on estuarine resources. The Peconic Estuary, Long Island, NY, USA, offers a naturally occurring nutrient loading gradient from eutrophic tidal creeks in the western region to mesotrophic bays in the eastern region. We conducted a five-month field grow-out experiment at four stations across this gradient to examine the effects of eutrophication on estuarine resources: eelgrasss (*Zostera marina*), juvenile bivalves (hard clams, *Mercenaria mercenaria*, Eastern oysters, *Crassostrea virginica*, and bay scallops, *Argopecten irradians*), and the non-resource suspension feeder, the slipper snail (*Crepidula fornicata*). Water quality parameters were also monitored at the field stations. Hard clams grew maximally at the least eutrophic site, but did not display a consistent east-west pattern along the eutrophication gradient. Eastern oysters grew maximally at the most eutrophic tidal creek, but also did not have a consistent east-west pattern. Bay scallops and slipper snails displayed consistent east-west patterns: growth was always maximal at the least eutrophic sites and minimal at the more eutrophic sites, although this pattern was more distinct for bay scallops. Eelgrass growth was depressed in the eutrophic tidal creek by low light levels and high epiphyte loading, maximal at a mid-estuary site, and depressed again at the less eutrophic site due to intense macro-benthic grazing. These results suggest that nutrient loading can have significant and complex effects on estuarine resource species, and that select species (e.g. oysters) may benefit from eutrophication under some conditions. Future ecosystem-based approaches that seek to restore estuarine

resources will need to account for the differential effects of nutrient loading as managers target species and regions to be restored.

Introduction:

Estuaries are home to a variety of valuable living resources. Finfish and shellfish are harvested directly in commercial and recreational fisheries, while seagrass beds are considered of paramount importance as structural habitat for shellfish and finfish in many coastal areas (Heck and Wetstone 1977, Irlandi and Peterson 1991, Beck *et al.* 2001). Many of the world's estuaries currently support lower abundances of finfish, shellfish, and seagrasses than they did historically due to overfishing (Jackson *et al.* 2001, Lotze *et al.* 2006), habitat loss (Orth *et al.* 2006), eutrophication (Nixon 1995, de Jonge *et al.* 2002), and harmful algal blooms (Hallegraeff 1993, Gobler *et al.* 2005, Sunda *et al.* 2006). As such, estuarine management plans are typically focused on combating these harmful processes and restoring living resources (Cloern 2001, Newell 2004, Lotze *et al.* 2006).

Changes in inorganic nutrient loading to estuaries can indirectly influence the growth of marine resource species. High rates of nutrient loading have been associated with increases in pelagic productivity, decreased water clarity, hypoxia, and declines in seagrass growth and abundances (Duarte 1995, Diaz and Rosenberg 2008, Wall *et al.* 2008), commonly referred to as the process of eutrophication (Nixon 1995, de Jonge et al. 2002). In response, estuarine management efforts often focus primarily on reducing anthropogenic nutrient loading in an effect to curb the negative effects of eutrophication (Cloern 2001, de Jonge *et al.* 2002). However, some level of nutrient loading must be necessary to sustain primary and secondary production (Nixon and Buckley 2002). High levels of inorganic nutrients favor larger phytoplankton cells (Malone 1980, Raven and Kubler 2002), such as diatoms and prymnesiophytes, which are generally considered a good source of nutrition for bivalves (Beukema and Cadee 1991, Wikfors *et al.* 1992, Weiss *et al.* 2007). Studies in several estuaries have shown blue mussels (*Mytilus*

edulis), hard clams (*Mercenaria mercenaria*), and softshell clams (*Mya arenaria*) can respond positively to increased nitrogen loading and high chlorophyll *a* levels in their habitats (van Stralen and Dijkema 1994, Weiss *et al.* 2002, Carmichael *et al.* 2004, Weiss *et al.* 2007). Weiss *et al.* (2002) and Carmichael *et al.* (2004) found that shell growth, soft tissue growth, and survival of *M. mercenaria* and *M. arenaria* increased along a naturally-occurring gradient of nitrogen loading in Waquoit Bay, Massachusetts, USA. They attribute these changes to increased quantity and quality of food particles due to nitrogen enrichment (Carmichael and Valiela 2005), although a similar response has not been found for bay scallops (*Argopecten irradians*, Shriver *et al.* 2002). While nutrient over-loading in estuaries has a well-known set of negative consequences (Valiela *et al.* 1992, Nixon 1995, Kemp *et al.* 2005), the stimulation of secondary production in bivalves could be an overlooked positive effect of nutrient loading (Nixon and Buckley 2002, Carmichael *et al.* 2004, Carmichael and Valiela 2005), especially in shallow ecosystems with well-mixed water columns that rarely experience hypoxia.

Where to locate the line between nutrient loading and nutrient **over**-loading remains an open question, and has clear ecological and financial implications for the management of estuaries (PEP-CCMP 2003). Clearly, the answer will be different for each estuary. Systems that are well-flushed will be able to absorb more anthropogenic nutrients without experiencing negative effects than systems that are poorly-flushed, stratified, or otherwise vulnerable to eutrophication (Cloern 2001, de Jonge et al. 2002). The biota of an estuary also influences how the system reacts to nutrient loading. The filtration capacity of an intact benthos, with bivalve suspension feeders, can buffer the negative effects of eutrophication (Officer et al. 1982, Smaal and Prins 1993, Cerco and Noel 2007). Some authors have suggested that aquacultured bivalves

could provide levels of ecosystem filtration comparable to wild populations (Newell 2004, Zhou et al. 2006, Huang et al. 2008).

The level at which nutrient loading and its effects becomes harmful to a particular living resource will also vary between species and with the overall health of the estuary. Seagrasses are sensitive to eutrophication and subsequent declines in light levels (Dennison et al. 1989, Duarte 1995), but could be recovered or facilitated by the presence of bivalves (Newell and Koch 2004, Wall et al. 2008, Carroll et al. 2008). Mobile fauna can avoid (at least temporarily) regions of low dissolved oxygen or poor water quality (Breitburg 2002), but most benthic organisms must simply endure the effects of eutrophication. Some species of bivalves, such as hard clams, soft-shell clams, and Eastern oysters (*Mercenaria mercenaria, Mya arenaria, Crassostrea virginica*) can endure short periods of hypoxia (Kraeuter and Castagna 2001, Carmichael et al. 2004, Kennedy et al. 1996) and may even benefit from nutrient-derived increases in quantity and quality of food particles (Weiss et al. 2002, Carmichael et al. 2004). Other bivalve species, such as bay scallops (*Argopecten irradians*) and blue mussels (*Mytilus edulis*) are more sensitive to declines in water quality, and may not survive the effects of high nutrient loads (Shriver et al. 2002, Altieri and Witman 2006). As plans for aquaculture and restoration of living resources proceed, it will be vital for managers to know how various levels of nutrient loading will affect the growth and survival of resource species (Cloern 2001, Newell 2004, Lotze et al. 2006). Ecosystem based-management (Slocombe 1993, Lotze et al. 2006) will also require a careful examination of where nutrient loads need to be reduced, where they can remain at current levels, and even where they might be allowed to increase somewhat (Nixon and Buckley 2002).

Eutrophication leads to shifts in distribution and abundance, and many potential nuisance organisms have increased their ranges and abundances in eutrophic systems, concurrent with the

removal of previously dominant organisms and the increase of nutrient loading (Mills 2001, Sunda et al. 2006, Lotze et al. 2006). The slipper shell *Crepidula fornicata* is a gastropod suspension feeder, is not currently a resource species, and is proliferating in areas formerly dominated by bivalve suspension feeders (Lewis et al. 1997, Lewis and Rivara 1998, Harke et al. in progress). While slipper shells may provide some of the filtration services formerly provided by bivalves (Barille et al. 2006, Harke et al. in progress), they are not likely to be viewed as a positive living resource in the same vein as clams, oysters, or scallops, and estuarine management plans should incorporate the response of potential nuisance species to changing levels of nutrient loading.

The Peconic Estuary, Long Island, NY, USA, is a system that may provide a unique opportunity to study the effects of nutrient loading on several marine resource species simultaneously. The Peconic Estuary is a chain of interconnected bays totaling 218 km^2 with an average depth of 4.7 m and an average tidal range of 0.76 m (Hardy 1976). There is a gradient of nutrient loading and chlorophyll *a* concentrations from high in the western regions (more concentrated human activities, less tidal mixing) to low in the eastern parts of the estuary (less concentrated human activities, more tidal mixing, Fig. 1, Hardy 1976). Hypoxia/anoxia is generally not present in the Peconic Estuary, due to wind and tidal mixing, and a lack of salinity-based stratification (Hardy 1976), so differences observed in the growth of estuarine resources should be due primarily to nutrient-loading, and not to hypoxia. The Peconic Estuary was formerly the site of productive fisheries for scallops, oysters, hard clams, soft-shell clams, eels, and menhaden (Hardy 1976), but these resources have been greatly diminished due to over-harvesting and the effects of the harmful brown tide algae (Cosper *et al.* 1987, Gobler *et al.*

2005). There have been concurrent losses of eelgrass habitat throughout the Peconic Estuary (Pickerell and Schott 2006).

This study was designed to examine the effects of nutrient loading across a naturally-occurring estuarine gradient on the growth and survival of estuarine resource species: juvenile hard clams (*M. mercenaria*), bay scallops (*A. irradians*), oysters (*C. virginica*), a juvenile planktivorous fish (sheepshead minnow, *Cyprinodon variegatus*), and eelgrass (*Z. marina*). Juvenile slipper shells (*C. fornicata*) were also included as a non-resource suspension feeder. These six species were placed at four field sites spaced across the nutrient-loading gradient. The growth of all populations along with levels of light, nutrients, size-fractionated chlorophyll *a*, and particulate organic matter were monitored during a growing season (Jun – Nov) to determine the effects of different levels of nutrient loading on the growth of estuarine resources.

Methods:

Four study sites were established in the Peconic Estuary across the estuarine gradient from a eutrophic tidal creek in western Flanders Bay to more oligotrophic conditions in Great and Little Peconic Bay (Fig 2). In order of west to east, and in order of most eutrophic to least eutrophic, the sites are named Meetinghouse Creek (MHC), Flanders Bay (FB), Great Peconic Bay (GPB), and Little Peconic Bay (LPB). Sites were approximately the same depth (1.8 – 2.5 m), and comparable in salinity and temperature, so the gradient of nutrient loading should be the primary factor driving differences in growth responses between the sites. The estuarine resources monitored at each site were eelgrass (*Zostera marina*), hard clams (*Mercenaria mercenaria*), eastern oysters (*Crassostrea virginica*), bay scallops (*Argopecten irradians*), and juvenile fish (sheepshead minnows, *Cyprinodon variegatus*). The growth of slipper shells

(*Crepidula fornicata*) was measured as a non-resource molluscan suspension feeder which has recently proliferated in areas formerly dominated by resource bivalves (Lewis et al. 1997, Lewis and Rivara 1998). Eelgrass shoots were placed in planters and lowered to the bottom, while juvenile fish and shellfish, obtained from a local hatchery, were placed in mesh cages (~2 mm mesh size) and bags kept in wire cages. Eelgrass planters and shellfish cages were anchored to the bottom, and a surface buoy marked each location. The growth of organisms at each study site was monitored bi-weekly over the course of the summer growing season from June to November 2008. Water quality parameters were also sampled bi-weekly such as chlorophyll *a*, dissolved oxygen, salinity, light attenuation, dissolved nutrient concentrations, and POC/PON concentrations. Temperature and ambient light levels were monitored continuously by HOBO© pendant-style data loggers. Differences in bulk water motion (tidal mixing + wind mixing) between sites were measured once using a plaster dissolution method (Doty 1971) averaged over a 24 hour period.

Chlorophyll *a* was measured in the whole and >5 μm size fractions using polycarbonate filters and standard fluorometric techniques (Parsons et al. 1984). Salinity and dissolved oxygen were measured using a YSI-### probe. Particulate organic carbon and particulate organic nitrogen were collected on pre-combusted glass fiber filters and processed using a CE Instruments Flash 1112 elemental analyzer (Sharp 1974, Gobler and Sañudo-Wilhelmy 2001).

Eelgrass was harvested and replaced every two weeks, and eelgrass productivity was measured by leaf area growth and above-ground biomass production (Ibarra-Obando and Boudouresque 1994). Epiphytes were scraped from eelgrass leaves for measurement of epiphyte loading. For each shellfish species, 50 individuals were stocked at each field site. These individuals were marked with bee-tags ("Queen Marking Kits," The Bee Works,

www.beeworks.com) attached with marine epoxy so individual growth rates by shell length could be measured. Clams and slipper shells were measured by shell length (anterior-posterior), while oysters and scallops were measured by shell height (hinge-ventral margin). Shell growth measurements were made every two weeks and at the end of the experiment and expressed as mean growth in mm $week^{-1}$. At the beginning of the experiment, 50 individuals of each species were sacrificed to obtain initial ash-free dry tissue weights (AFDW) and condition indices. Ash-free dry tissue weights and condition indices were measured on all surviving individuals at the end of the experiment, and mean growth rates by weight were calculated in mg AFDW $week^{-1}$. Condition indices were calculated according to the methods of Rheault and Rice (1996). Juvenile sheepshead minnows were collected and replaced every two weeks. Sheepshead minnow growth was measured in mm $week^{-1}$ and mg dry wt $week^{-1}$. Length-weight regressions were used to estimate initial weights for sheepshead minnows. Shellfish and cages were cleaned of fouling material every two weeks, while fish cages were replaced every two weeks.

Growth of fish, shellfish, and eelgrass was analyzed using one- and two-way ANOVAs to check for significant differences between sites and time. A two-way repeated measures ANOVA was used to analyze measurements of tagged individuals over several time points. When data did not fit the ANOVA assumptions, a non-parametric test was used (Kruskal-Wallis test). Correlation and regression analyses were used to account for growth responses in terms of water quality parameters. Chlorophyll *a* and light level trends were analyzed using two-way ANOVAs with site and time-point. When a significant effect on the response variables was detected, multiple comparison tests (Tukey's Studentized range) was used to test for significant differences between levels within the treatment. Mortality of juvenile bivalves was analyzed

using a G-test of independence (Sokal and Rohlf 1995). All statistical results were considered significant if $p < 0.05$. Statistical analyses were carried out on the software SigmaStat 3.0.

Results:

Field sampling was carried out bi-weekly from June 30th, 2008, to November 5th, 2008. Physical trends of temperature, salinity, and dissolved oxygen for bottom waters (depth of ~ 2 m) are shown in Fig 3A-C. Temperatures of bottom waters started off at 25 – 27° C in July and declined to 10 – 11° C by early November, with only small difference between sites (Fig 3 A). Salinities of bottom waters were consistent over the course of the season, but did differ slightly between sites, ranging from 26 – 27 psu at Meetinghouse Creek, the most inland site, to 28 – 29 psu at Great and Little Peconic Bays, the sites closest to the ocean exchange (Fig 3 B). Dissolved oxygen in bottom waters differed more dramatically by both date and site, with bottom waters of Meetinghouse Creek often hypoxic or nearly anoxic during July – September (Fig 3 C). Dissolved oxygen varied at the other three sites but remained normoxic throughout the study (Fig 3 C). Since we anticipated hypoxia in the bottom waters of Meetinghouse Creek, the shellfish cages were placed at a depth of ~ 1 m, which should have avoided the hypoxia at 2 m.

Chlorophyll *a* in the whole and >5 μm size differed significantly by site, by date, and by site x date interaction ($p<0.001$, 2-way ANOVA, Fig 4 A). Meetinghouse Creek consistently had high chl *a*, 20 - 40 μg L^{-1}, while Flanders Bay had intermediate chl *a*, 5 – 15 μg L^{-1}, and Great and Little Peconic Bays had low chl *a*, <10 μg L^{-1} all season. Even though there was temporal variation in chl *a* at all sites, this trend of high phytoplankton biomass in the west and lower phytoplankton biomass in the east is consistent when values for each site are averaged

across time points ($p<0.001$, 1-way ANOVA, Fig 4 B). Virtually all of the chl *a* was in the >5 μm size fraction at Meetinghouse Creek, and the proportion of phytoplankton in the larger size fraction decreased from west to east, but was, on average, never less than 76% of the total phytoplankton community (Fig 4 C).

The growth of all shellfish was generally high during the summer and declined with decreasing temperatures in October and November (Figs 5-7 C-D). The growth of hard clams (*Mercenaria mercenaria*) averaged over the whole growing season was significantly different between sites, when measured by shell growth ($p<0.001$, ANOVA, Fig 5-A) or by soft-tissue growth ($p<0.001$, ANOVA-on-ranks, Fig 5-B). By both measures, the clams at L. Peconic B. grew fastest, but there was not a consistent east-west pattern along the eutrophication gradient. Hard clams in Mtghouse Cr. had the second-fastest growth by shell length or AFDW, with clams at the mid-estuarine sites, Flanders B. and Gr. Peconic B., growing more slowly (Fig 5 A-B). There was also significant variation by date and a site x date interaction for hard clams ($p<0.001$, 2-way repeated measures ANOVA, Fig 5 C-D). For example, shell growth of hard clams was slow at Mtghouse Cr. in the mid-summer and accelerated in the early fall (Fig 5 C-D).

The growth of eastern oysters (*Crassostrea virginica*) averaged over the whole growing season was also significantly different between sites, when measured by shell growth ($p<0.001$, ANOVA-on-ranks, Fig 6-A) or by soft-tissue growth ($p<0.001$, ANOVA-on-ranks, Fig 6-B). By both measures, the oysters at Mtghouse Cr. grew fastest, but as with the hard clams, there was not a consistent east-west pattern along the eutrophication gradient. Eastern oysters in L. Peconic B. had the second fastest growth by shell length or AFDW, with oysters in the middle sites of Flanders B. and Gr. Peconic B. growing more slowly (Fig 6 A-B). The variation in shell growth rates of tagged individuals was significant by date and site x date interaction ($p<0.001$, 2-

way repeated measures ANOVA, Fig 6 C-D), but there was not a significant difference by site. Nevertheless, the shell growth of oysters in Mtghouse Cr. had a similar pattern to clam growth at this site: oyster growth was slow in mid-summer and accelerated greatly in early fall (Fig 6 C-D).

The growth of bay scallops (*Argopecten irradians*) averaged over the whole growing season was significantly different by site when measured by shell growth ($p<0.001$, ANOVA, Fig 7-A) or by soft-tissue growth ($p<0.001$, ANOVA, Fig 7-B). In contrast to the hard clams and eastern oysters, bay scallops had a growth response with a consistent east-west pattern: scallop growth was highest in the east, at the least eutrophic site, and decreased stepwise to the west at the more eutrophic sites (Fig 7 A-B). Shell growth by date follows the same pattern, with a clear separation in mean shell height between the eastern sites (less eutrophic) and the western sites (more eutrophic) throughout the season (Fig 7 C-D). Shell growth was significantly different by site, date, and site x date interaction ($p<0.001$, 2-way repeated measures ANOVA, Fig 7 C-D).

The mortality of shellfish was not significantly different by site for all three species (G-test of independence), however the percent survival by site tended to follow the growth trends by site, i.e. sites with the slowest growth had the highest mortality for that species and vice versa (data not shown). The growth of juvenile fish (sheepshead minnows, *Cyprinodon variegatus*) was not significantly different by site (1- and 2-way ANOVAs, data not shown).

Bulk water motion, as measured by rates of plaster dissolution, was significantly greater in the east, and decreased stepwise to the west, with water motion at all sites also significantly greater than a "no-flow" control ($p<0.001$, ANOVA, Fig 8). There was no significant difference in bulk water motion between plaster blocks placed inside shellfish cages and outside shellfish cages (2-way ANOVA). However, we were not able to place plaster blocks inside the individual

shellfish bags, which were 2-mm mesh and may have reduced water motion relative to outside the mesh.

The growth of slipper snails (*Crepidula fornciata*) was significantly different between sites when averaged over the whole season for shell growth (Fig 9A) or soft tissue growth (Fig 9B). The shell growth of slipper snails (Fig 9A) paralleled that of bay scallops (Fig 7A); slipper snail growth was significantly higher at the eastern sites compared to Flanders Bay and Meetinghouse Creek ($p<0.001$, 1-way ANOVA). Oddly, the soft tissue growth and shell growth displayed differing patterns for slipper snails (Fig 9B). Individuals at Great Peconic Bay that had the fastest shell growth had relatively low soft tissue growth (Fig 9A-B, $p<0.05$, 1-way ANOVA). Slipper snails in Meetinghouse Creek showed the slowest growth, by shell length or by soft tissue weight (Fig 9A-B).

The growth of eelgrass (*Zostera marina*) was measured on a per shoot basis by leaf area productivity (Fig 10A) and mass productivity (Fig 10B). For both measures, there was a depression in growth from July-early August when temperatures were warmest, a peak in growth from late August-September, and another depression in growth at the end of the season as temperatures cooled (Fig 10A-B). Leaf area productivity of eelgrass shoots was significantly higher at Flanders Bay, a mid-estuary site (Fig 10A, $p<0.01$, 2-way ANOVA), and this effect was most pronounced from late August-September. Mass productivity of eelgrass shoots (Fig 10B) followed a similar pattern, although the trend was not significant. The density of epiphytes on eelgrass blades was consistently higher at Meetinghouse Creek than the other sites from July-September (Fig 11, $p<0.01$, ANOVA on ranks), and then declined at the end of the season. The eastern estuary sites (Great and Little Peconic Bays) had a strong and persistent disturbance of eelgrass shoots from macro-benthic animals, notably spider crabs *Libinia emarginata* (Fig 12,

pers. obs.). The number of eelgrass shoots uprooted by these animals was significantly higher at the eastern sites throughout the season (Fig 12, $p<0.01$, 2-way ANOVA). In addition, many of the eelgrass shoots that remained rooted at these sites also showed evidence of grazing (data not shown).

Discussion:

Over the course of a growing season (Jun-Nov 2008), we demonstrated that the growth of eelgrass and four species of juvenile shellfish differed significantly at sites along an eutrophication gradient in the Peconic Estuary, NY, USA. The trend for three bivalve species was the same whether measured by shell growth or soft tissue growth, while the growth patterns of the gastropod *Crepidula fornicata* differed between shell growth and soft tissue growth. The growth responses for each species indicate that some estuarine resources had a consistent response to changing levels of nutrient loading and chlorophyll *a* (e.g. bay scallops), while other species had complex and possibly multi-modal response to a range of nutrient loading levels (e.g. hard clams, eastern oysters, eelgrass).

None of the shellfish species studied appeared to be solely food-limited, or at least not limited by food as measured by whole or > 5 μm chlorophyll *a*. The growth of juvenile hard clams (*M. mercenaria*) and eastern oysters (*C. virginica*) both displayed a "bi-modal" response to the eutrophication gradient of the Peconic Estuary. Both species had maximal growth at the two ends of the estuary, and minimal growth at the stations in between (Fig. 5 A-B, 6 A-B). High growth rates at the eutrophic end of the estuary (Mtghouse Cr.) could be due to an increase in the quantity and quality of seston under high-nutrient loading conditions (Carmichael et al. 2004, Carmichael and Valiela 2005). High growth rates at the oligotrophic end of the estuary

could be due to increased flow from wind and tidal mixing (Fig 8), combined with more stable temperatures, salinities, and dissolved oxygen levels. It is difficult to explain the slower growth at the intermediate stations, especially Gr. Peconic B., where temperature, salinity, DO, and chl *a* (Fig 3 A-C, Fig 4 A-C) were indistinguishable from L. Peconic B., which promoted fast growth for all three species. Bulk water motion was significantly greater at L. Peconic B., and under oligotrophic conditions, this variable may have made the greatest difference to hard clams and scallops, and secondarily to oysters (Kennedy et al. 1996). Oysters are known to feed at faster rates than other shellfish (Tenore and Dunstan 1973) and may have been better able to take advantage of very dense phytoplankton communities in Mtghouse Cr. than the other shellfish species.

This study is consistent with the finding that bay scallops are generally not food-limited in most estuaries (Shriver et al. 2002). Scallops grew maximally at the lowest chl *a*, highest flow sites and minimally in the most eutrophic sites (Fig 7 A-B). This trend was maintained across the whole season, with less site x date variation than the other species (Fig 7 C-D). Although the trends in scallop mortality were not significant, the slowest-growing scallops in Mtghouse Cr. also had the highest mortality, while the faster growing scallops in Gr. and L. Peconic Bays had the lowest mortality.

The shell growth of slipper snails paralleled the growth of bay scallops (Fig 7A-B, Fig 9A), although the geographic trend was more pronounced for bay scallops (Fig 7B). This suggests that both species would perform well in the relatively oligotrophic, high-flow, low chlorophyll eastern regions of the estuary, and that there may be competition or niche over-lap between bay scallops and slipper snails. Recent reports have suggested that slipper snails are proliferating in areas of the Peconic Estuary formerly dominated by bay scallops (Lewis et al.

1997, Lewis and Rivara 1998) and that slipper snails are able to filter phytoplankton communities containing very small or harmful algal cells more efficiently than bivalve suspension feeders (Kach and Ward 2008, Harke et al. in progress).

Due to previous findings on the effects of eutrophication on eelgrass (Dennison et al. 1989, Duarte 1995, Wall et al. 2008), we expected the growth of eelgrass (*Z. marina*) to be maximal at the eastern end of the estuary, which has relatively high light levels and low phytoplankton biomass, and minimal in the dim light of the eutrophic tidal creek. Instead, eelgrass growth has a distinct "mid-estuary peak" at the Flanders Bay site (Fig 10A-B), and this effect is most prominent from late August through September. It is likely that low light levels (data not shown) and high epiphyte loading (Fig 11) slowed the growth of eelgrass in the most eutrophic site, Meetinghouse Cr. But even though light levels were higher at the eastern end of the estuary (data not shown), there was strong, persistent grazing disturbance at Great and Little Peconic Bays that approached 80% of all eelgrass shoots uprooted (Fig 12). The grazing and uprooting of eelgrass was mostly due to spider crabs, *Libinia emarginata* (pers. obs.). As such, the mid-estuary site, Flanders Bay, with intermediate light levels and relatively little grazing disturbance, seemed to represent the optimum site for eelgrass growth in this study. However, it is likely that eelgrass shoots that were adequately caged or shielded from grazers would perform well under the higher light levels at the eastern end of the estuary.

Our goal was to isolate the effects of nutrient-loading on bivalve growth. Although we tried to choose sites that were as similar as possible in physical characteristics, there were differences in this regard. Mtghouse Cr., the tidal creek with high nutrients and high chl *a*, was also more hypoxic, and somewhat warmer than the other three sites (Fig 3 A-C). This site also experienced a toxic algal bloom during late August and September (*Cochlodinium polykrikoides*;

data not shown). These variables, along with bulk water motion, represent possible confounding variables in our nutrient-loading study. Mid-summer temperatures at Mtghouse Cr. Were likely too warm (27-28° C, Fig 3 A) for maximal hard clam growth (Kraeuter and Castagna 2001; Weiss et al 2007); hard clam growth at this site later accelerated when temperatures dropped below 27° C (Fig 5 D). Interestingly, hard clams and oysters displayed their seasonal maximal and near-maximal growth rates, respectively, for all sites in Mtghouse Cr. during mid-September when bottom oxygen levels rose out of the hypoxic range and the *C. polykrikoides* bloom subsided. This finding suggests that while the food supply at this site was ideal for these two species, class symptoms of eutrophication such as low bottom oxygen levels and harmful algal blooms prohibited these shellfish from benefiting from an abundant food supply. Such interactions of multiple physical and biological variables making growth rate comparisons based on nutrient loading and chlorophyll *a* difficult. Eventually a multi-variable regression model for the growth of juveniles of each species will be constructed and compared to growth models for eelgrass (*Zostera marina*), another critical restoration species. These models may help managers to target proposed restoration species to the areas of an estuary where they will have the greatest chance of success.

Many anthropogenic insults have led to the degradation of estuaries, with the concurrent loss of estuarine resources (Lotze et al. 2006) such as clams, oysters, scallops, and eelgrass. Increases in anthropogenic nutrient loading, leading to eutrophication, have been identified as a primary driver of estuarine degradation (Nixon 1995, de Jonge et al. 2002), but recent research and reviews have raised the possibility of beneficial nutrient loading under some conditions and for some species (Nixon and Buckley 2002, Carmichael et al. 2004). Undoubtedly, eutrophication is a complex problem that continues to challenge our scientific understanding, and

will require a variety of management approaches (Cloern 2001). Jackson (2001) and Lotze et al. (2006) have emphasized the need to restore upper trophic levels and suspension feeders as effective top-down controls on estuarine ecosystems. The re-seeding or restoration of suspension feeding bivalves in estuaries can have multiple benefits: the bivalves themselves are a resource for commercial and recreational fisheries, while simultaneously providing ecosystem services of filtration (Newell 1988, Dame 1996, Altieri and Witman 2006), top-down control of algal blooms (Officer et al. 1982, Cerrato et al. 2004), and transfer of nutrients and biomass to the benthos (Smaal and Prins 1993, Newell 2004). It is possible that bivalve aquaculture can achieve many of these goals simultaneously (Newell 2004, Huang et al. 2008), although over-stocking of aquaculture operations can have negative effects on adjacent wild and cultured bivalves (Newell 2004, Zhou et al. 2006).

Seagrass habitats are disappearing at an alarming rate worldwide (Orth et al. 2006), and restoration of seagrass beds could provide valuable habitat for fin fish and shellfish (Irlandi and Peterson 1991, Beck et al. 2001). The simultaneous restoration and management of seagrass beds and bivalve populations will likely have synergistic effects (Wall et al. 2008, Carroll et al. 2008), as each group of organisms provides multiple benefits for the other.

Any plan to re-seed or restore seagrasses or bivalve populations into a eutrophic system will need to take into account the species in question and its responses to varying levels of nutrient loading. Based on our findings, we can recommend that restoration of scallops will have the greatest chance of success in high flow environments that are relatively oligotrophic, although inter-specific competition with *C. fornicata*, which also grew maximally in this region, could affect *A. irradians* growth. Restoration of clams and oysters could proceed over a wider variety of estuarine habitats, with oysters should being a prime candidate for restoration in the

most impacted, eutrophic estuarine waters. Finally, eelgrass restoration sites must balance light and nutrient regimes with the activity of benthic grazers which may disrupt eelgrass growth.

Acknowledgements:

Many people provided field and laboratory support for this project, including C. Blankenship, R. Goodale, B. Rodgers, A. Burson, M. Marcoval, S. Ingles, J. Goleski, T. Duffy, J. Carroll, M. Harke, T. Hattenrath, T. Davis, A. Ehmer, T. Behling, K. Montanye, K. Lavelle, M. Meade, D. Getz, C. MacIntyre, and B. Gagliardi. Juvenile shellfish were provided by G. Rivara and M. Patricio from Cornell Cooperative Extension, Southold, NY. Access to Meetinghouse Creek field site was granted by Larry's Lighthouse Marina, Aquebogue, NY. Thanks to Evan Ward for advice on tagging shellfish. This project was supported by funding from The Nature Conservancy.

References:

Altieri AH, Witman JD (2006) Local extinction of a foundation species in a hypoxic estuary: Integrating individuals to ecosystem. Ecology 87:717-730

Barille L, Cognie B, Beninger P, Decottignies P, Rince Y (2006) Feeding responses of the gastropod Crepidula fornicata to changes in seston concentration. Marine Ecology-Progress Series 322:169-178

Beck MW, Heck KL, Able KW, Childers DL, Eggleston DB, Gillanders BM, Halpern B, Hays CG, Hoshino K, Minello TJ, Orth RJ, Sheridan PF, Weinstein MR (2001) The identification, conservation, and management of estuarine and marine nurseries for fish and invertebrates. Bioscience 51:633-641

Beukema JJ, Cadee GC (1991) Growth-Rates of the Bivalve Macoma-Balthica in the Wadden Sea During a Period of Eutrophication - Relationships with Concentrations of Pelagic Diatoms and Flagellates. Marine Ecology-Progress Series 68:249-256

Breitburg D (2002) Effects of hypoxia, and the balance between hypoxia and enrichment, on coastal fishes and fisheries. Estuaries 25:767-781

Carmichael RH, Shriver AC, Valiela I (2004) Changes in shell and soft tissue growth, tissue composition, and survival of quahogs, Mercenaria mercenaria, and softshell clams, Mya arenaria, in response to eutrophic-driven changes in food supply and habitat. Journal of Experimental Marine Biology and Ecology 313:75-104

Carmichael RH, Valiela L (2005) Coupling of near-bottom seston and surface sediment composition: Changes with nutrient enrichment and implications for estuarine food supply and biogeochemical processing. Limnology and Oceanography 50:97-105

Carroll J, Gobler CJ, Peterson BJ (2008) Resource-restricted growth of eelgrass in New York estuaries: light limitation, and alleviation of nutrient stress by hard clams. Marine Ecology-Progress Series 369:51-62

Cerco CF, Noel MR (2007) Can oyster restoration reverse cultural eutrophication in Chesapeake Bay? Estuaries and Coasts 30:331-343

Cloern JE (2001) Our evolving conceptual model of the coastal eutrophication problem. Marine Ecology-Progress Series 210:223-253

Cosper EM, Dennison WC, Carpenter EJ, Bricelj VM, Mitchell JG, Kuenstner SH, Colflesh D, Dewey M (1987) Recurrent and Persistent Brown Tide Blooms Perturb Coastal Marine Ecosystem. Estuaries 10:284-290

de Jonge VN, Elliott M, Orive E (2002) Causes, historical development, effects and future challenges of a common environmental problem: eutrophication. Hydrobiologia 475:1-19

Dame RF (1996) Ecology of marine bivalves: an ecosystem approach. CRC Press, Boca Raton, FL

Dennison WC, G.J. Marshall, C. Wigand (1989) Effects of "brown tide" shading on eelgrass (*Zostera marina*) distributions, Vol. Springer, New York

Diaz RJ, Rosenberg R (2008) Spreading dead zones and consequences for marine ecosystems. Science 321:926-929

Doty MS (1971) Measurement of Water Movement in Reference to Benthic Algal Growth. Bot Marina 14:32-&

Duarte CM (1995) Submerged Aquatic Vegetation in Relation to Different Nutrient Regimes. Ophelia 41:87-112

Gobler CJ, Lonsdale DJ, Boyer GL (2005) A review of the causes, effects, and potential management of harmful brown tide blooms caused by Aureococcus anophagefferens (Hargraves et Sieburth). Estuaries 28:726-749

Gobler CJ, Sanudo-Wilhelmy SA (2001) Temporal variability of groundwater seepage and brown tide blooms in a Long Island embayment. Marine Ecology-Progress Series 217:299-309

Hallegraeff GM (1993) A Review of Harmful Algal Blooms and Their Apparent Global Increase. Phycologia 32:79-99

Hardy CD (1976) A preliminary description of the Peconic Bay Estuary. Report No. 76-4, Marine Sciences Research Center, Stony Brook, NY

Heck KL, Wetstone GS (1977) Habitat Complexity and Invertebrate Species Richness and Abundance in Tropical Seagrass Meadows. Journal of Biogeography 4:135-142

Huang CH, Lin HJ, Huang TC, Su HM, Hung JJ (2008) Responses of phytoplankton and periphyton to system-scale removal of oyster-culture racks from a eutrophic tropical lagoon. Marine Ecology-Progress Series 358:1-12

Ibarraobando SE, Boudouresque CF (1994) An Improvement of the Zieman Leaf Marking Technique for Zostera-Marina Growth and Production Assessment. Aquat Bot 47:293-302

Irlandi EA, Peterson CH (1991) Modification of Animal Habitat by Large Plants - Mechanisms by Which Seagrasses Influence Clam Growth. Oecologia 87:307-318

Jackson JBC (2001) What was natural in the coastal oceans? Proceedings of the National Academy of Sciences of the United States of America 98:5411-5418

Jackson JBC, Kirby MX, Berger WH, Bjorndal KA, Botsford LW, Bourque BJ, Bradbury RH, Cooke R, Erlandson J, Estes JA, Hughes TP, Kidwell S, Lange CB, Lenihan HS, Pandolfi JM, Peterson CH, Steneck RS, Tegner MJ, Warner RR (2001) Historical overfishing and the recent collapse of coastal ecosystems. Science 293:629-638

Kach, D. J. and J. E. Ward (2008). "The role of marine aggregates in the ingestion of picoplankton-size particles by suspension-feeding molluscs." Marine Biology **153**(5): 797-805.

Kemp WM, Boynton WR, Adolf JE, Boesch DF, Boicourt WC, Brush G, Cornwell JC, Fisher TR, Glibert PM, Hagy JD, Harding LW, Houde ED, Kimmel DG, Miller WD, Newell RIE, Roman MR, Smith EM, Stevenson JC (2005) Eutrophication of Chesapeake Bay: historical trends and ecological interactions. Marine Ecology-Progress Series 303:1-29

Kennedy VS, Newell RIE, Eble AF (1996) The eastern oyster *Crassostrea virginica*, Maryland Sea Grant, College Park, MD

Kraeuter JN, Castagna M (2001) Biology of the Hard Clam, Vol. Elsevier Science, New York

Lewis D, Kassner J, Cerrato R, Finch R (1997) An Assessment of Shellfish Resources in the Deep Water Areas of the Peconic Estuary. Marine Sciences Research Center, State University of New York, Stony Brook, N.Y. pp 17, F-92, F-96

Lewis DE, Rivara G (1998) An Assessment of Shellfish Resources in the Tributaries and Embayments of the Peconic Estuary. Cornell Cooperative Extension of Suffolk County Marine Program, Riverhead, NY pp 20

Lotze HK, Lenihan HS, Bourque BJ, Bradbury RH, Cooke RG, Kay MC, Kidwell SM, Kirby MX, Peterson CH, Jackson JBC (2006) Depletion, degradation, and recovery potential of estuaries and coastal seas. Science 312:1806-1809

Malone TC (1980) Algal size. In: Morris I (ed) Thy Physiological Ecology of Phytoplankton. Blackwell, Oxford, p 433-463

Mills CE (2001) Jellyfish blooms: are populations increasing globally in response to changing ocean conditions? Hydrobiologia 451:55-68

Newell RIE (2004) Ecosystem influences of natural and cultivated populations of suspension-feeding bivalve molluscs: A review. J Shellfish Res 23:51-61

Newell RIE, Koch EW (2004) Modeling seagrass density and distribution in response to changes in turbidity stemming from bivalve filtration and seagrass sediment stabilization. Estuaries 27:793-806

Nixon SW (1995) Coastal Marine Eutrophication - a Definition, Social Causes, and Future Concerns. Ophelia 41:199-219

Nixon SW, Buckley BA (2002) "A strikingly rich zone" - Nutrient enrichment and secondary production in coastal marine ecosystems. Estuaries 25:782-796

Officer CB, Smayda TJ, Mann R (1982) Benthic Filter Feeding - a Natural Eutrophication Control. Marine Ecology-Progress Series 9:203-210

Orth RJ, Carruthers TJB, Dennison WC, Duarte CM, Fourqurean JW, Heck KL, Hughes AR, Kendrick GA, Kenworthy WJ, Olyarnik S, Short FT, Waycott M, Williams SL (2006) A global crisis for seagrass ecosystems. Bioscience 56:987-996

PEPCCMP (2003) Peconic Estuary Program Comprehensive Conservation and Management Plan.

Parsons TR, Y. Maita, C.M. Lalli (1984) A manual of chemical and biological methods for seawater analysis., Vol. Pergamon Press, Oxford
Pickerell C, Schott S (2006) Peconic Estuary Program eelgrass (*Zostera marina*) long term monitoring project. Peconic Estuary Program, Suffolk County Office of Ecology, Yaphank, NY
Raven JA, Kubler JE (2002) New light on the scaling of metabolic rate with the size of algae. Journal of Phycology 38:11-16
Rheault RB, Rice MA (1996) Food-limited growth and condition index in the eastern oyster, Crassostrea virginica (Gmelin 1791), and the bay scallop, Argopecten irradians irradians (Lamarck 1819). J Shellfish Res 15:271-283
SCDHS (1976-2005) Annual report on water quality in Suffolk County's coastal waters. Suffolk County (NY) Department of Health Services, Office of Ecology
Sharp JH (1974) Improved Analysis for Particulate Organic Carbon and Nitrogen from Seawater. Limnology and Oceanography 19:984-989
Shriver AC, Carmichael RH, Valiela I (2002) Growth, condition, reproductive potential, and mortality of bay scallops, Argopecten irradians, in response to eutrophic-driven changes in food resources. Journal of Experimental Marine Biology and Ecology 279:21-40
Slocombe DS (1993) Implementing Ecosystem-Based Management. Bioscience 43:612-622
Smaal AC, Prins TC (1993) The uptake of organic matter and the release of inorganic nutrients by bivalve suspension feeder beds. In: Dame RF (ed) Bivalve filter feeders in estuarine and coastal ecosystem processes. Springer- Verlag, Berlin, p 271–298
Sokal RR, Rohlf FJ (1995) Biometry. WH Freeman, New York
Sunda WG, Graneli E, Gobler CJ (2006) Positive feedback and the development and persistence of ecosystem disruptive algal blooms. Journal of Phycology 42:963-974
Tenore KR, Dunstan WM (1973) Comparison of Feeding and Biodeposition of 3 Bivalves at Different Food Levels. Mar Biol 21:190-195
Valiela I, Foreman K, Lamontagne M, Hersh D, Costa J, Peckol P, Demeoandreson B, Davanzo C, Babione M, Sham CH, Brawley J, Lajtha K (1992) Couplings of Watersheds and Coastal Waters - Sources and Consequences of Nutrient Enrichment in Waquoit Bay, Massachusetts. Estuaries 15:443-457
Vanstralen MR, Dijkema RD (1994) Mussel Culture in a Changing Environment - the Effects of a Coastal Engineering Project on Mussel Culture (Mytilus-Edulis-L) in the Oosterschelde Estuary (Sw Netherlands). Hydrobiologia 283:359-379
Wall CC, Peterson BJ, Gobler CJ (2008) Facilitation of seagrass Zostera marina productivity by suspension-feeding bivalves. Marine Ecology-Progress Series 357:165-174
Weiss ET, Carmichael RH, Valiela I (2002) The effect of nitrogen loading on the growth rates of quahogs (Mercenaria mercenaria) and soft-shell clams (Mya arenaria) through changes in food supply. Aquaculture 211:275-289
Weiss MB, Curran PB, Peterson BJ, Gobler CJ (2007) The influence of plankton composition and water quality on hard clam (Mercenaria mercenaria L.) populations across Long Island's south shore lagoon estuaries (New York, USA). Journal of Experimental Marine Biology and Ecology 345:12-25
Wikfors GH, Ferris GE, Smith BC (1992) The Relationship between Gross Biochemical-Composition of Cultured Algal Foods and Growth of the Hard Clam, Mercenaria-Mercenaria (L). Aquaculture 108:135-154

Zhou Y, Yang HS, Zhang T, Liu SL, Zhang SM, Liu Q, Xiang JH, Zhang FS (2006) Influence of filtering and biodeposition by the cultured scallop Chlamys farreri on benthic-pelagic coupling in a eutrophic bay in China. Marine Ecology-Progress Series 317:127-141

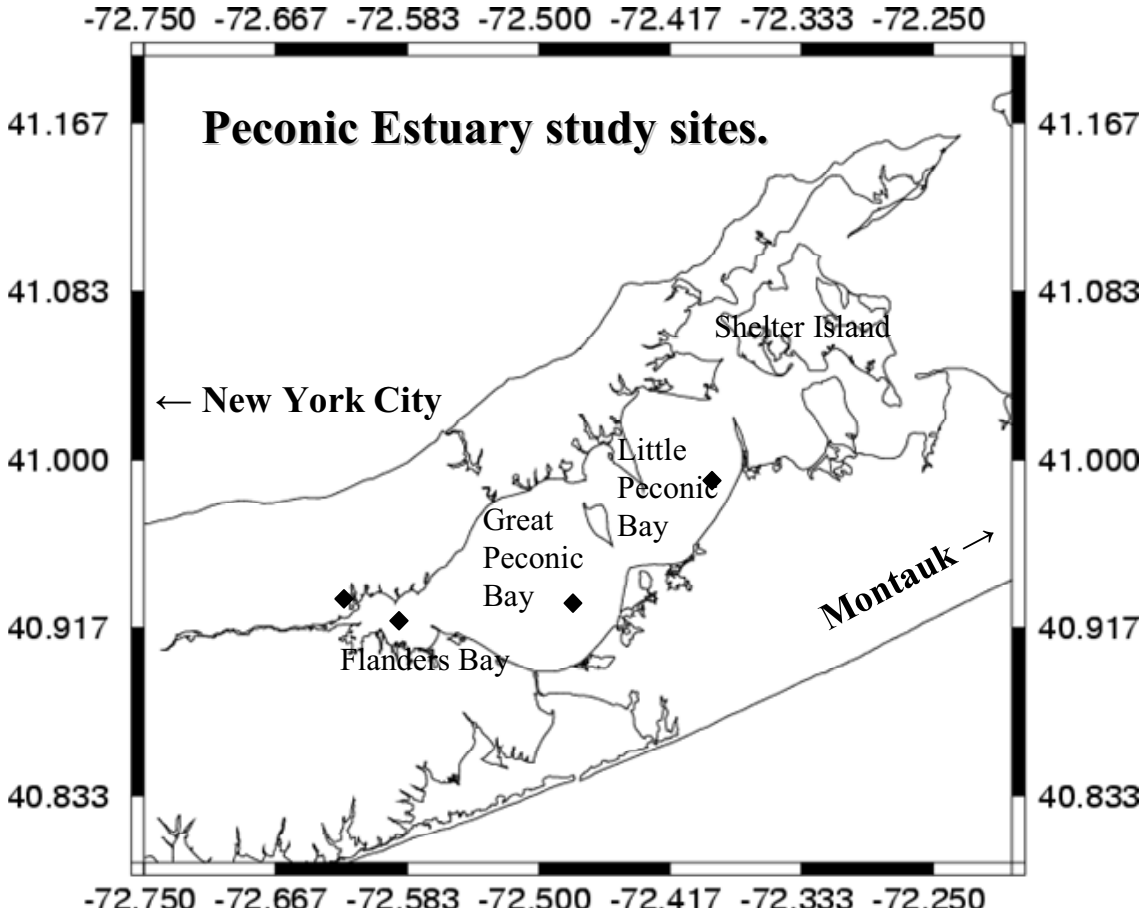


Figure 1. Peconic Estuary study sites marked by: ◆

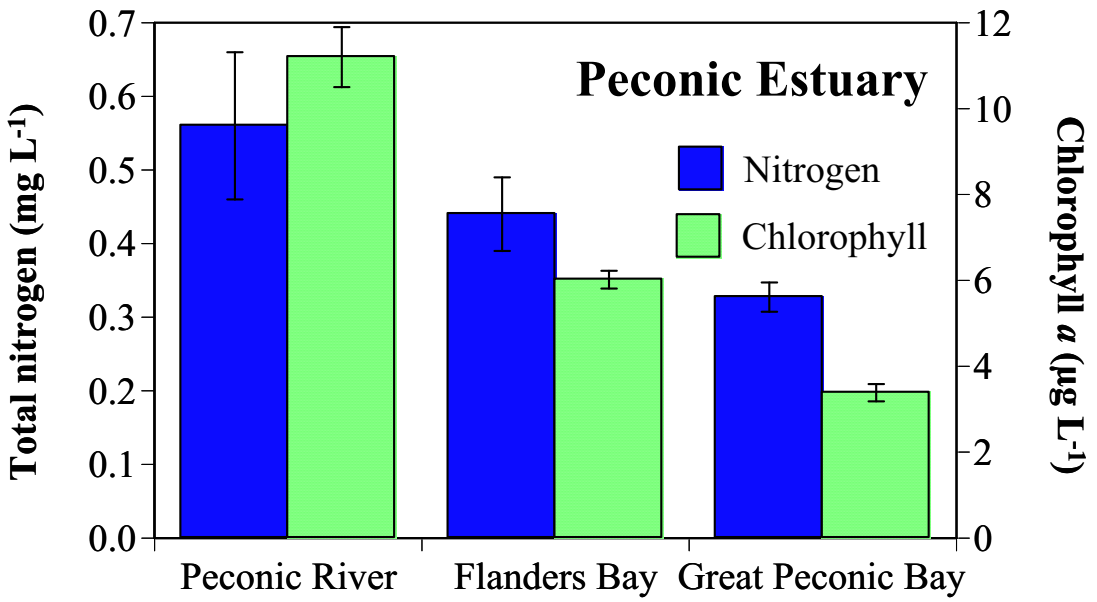
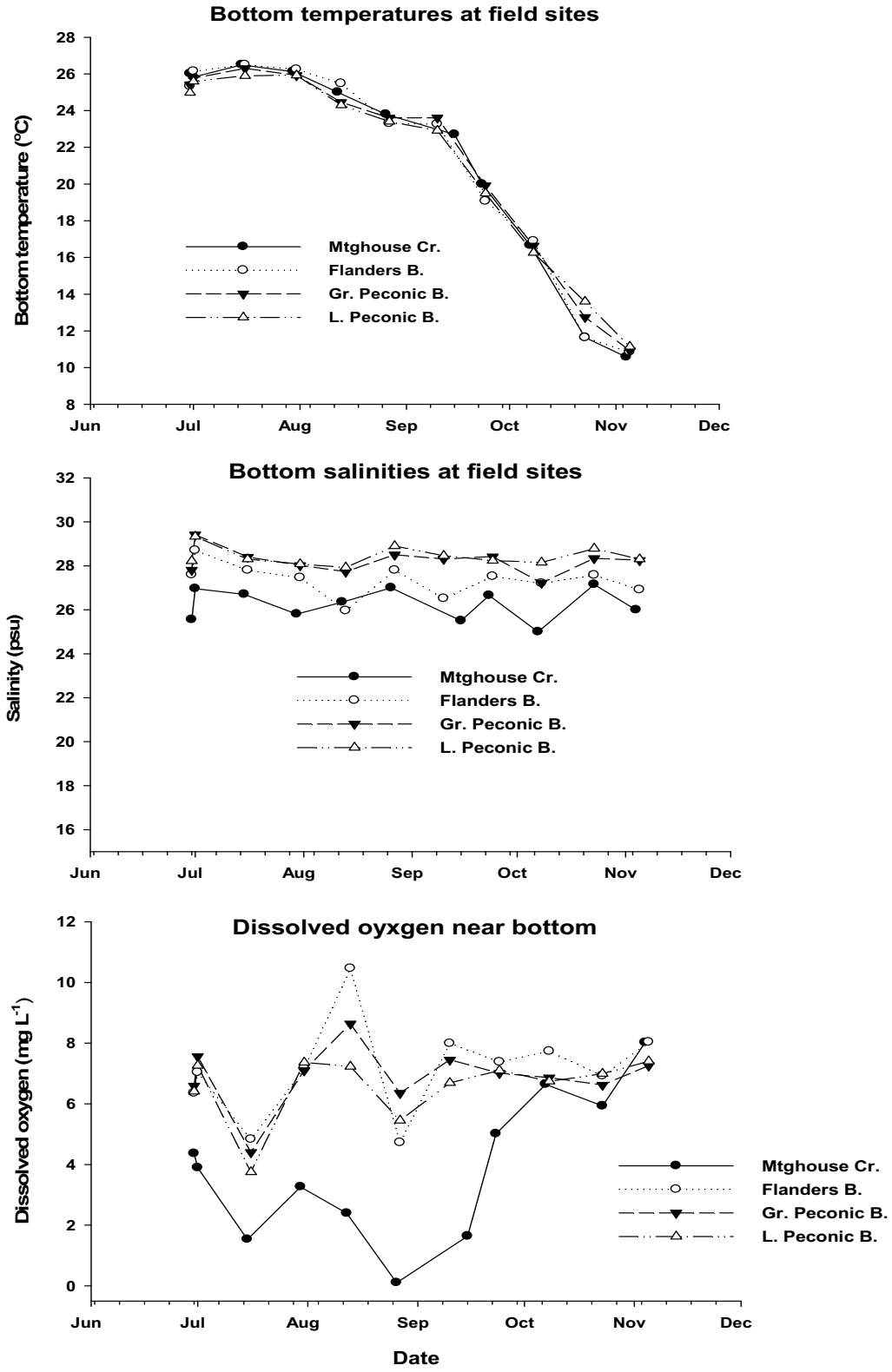


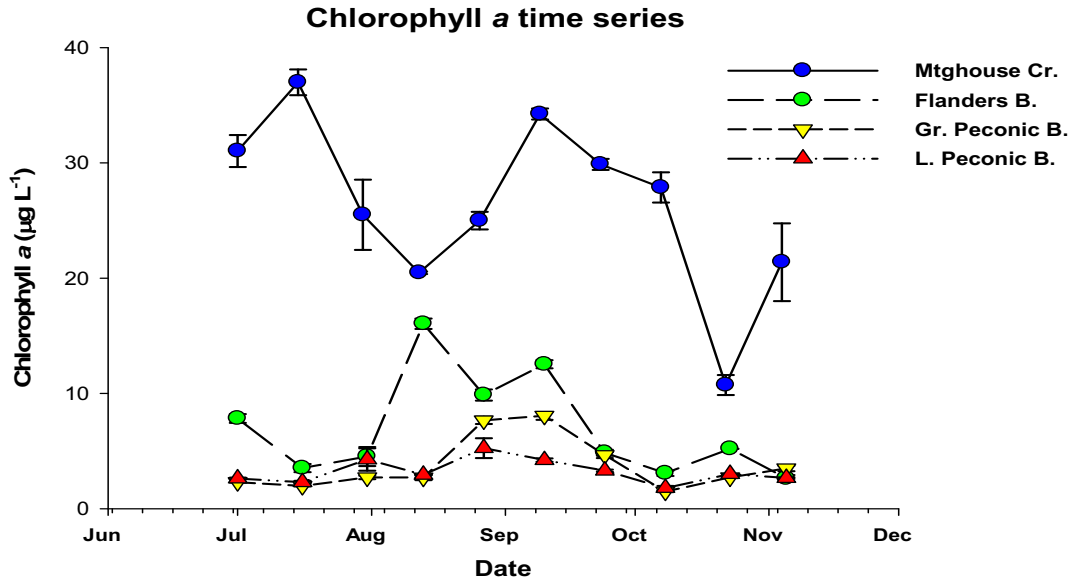
Figure 2. Concentrations of chlorophyll *a* and total nitrogen across the study area in the western Peconic estuary (stations 240, 110, 130; SCDHS 1976-2005).

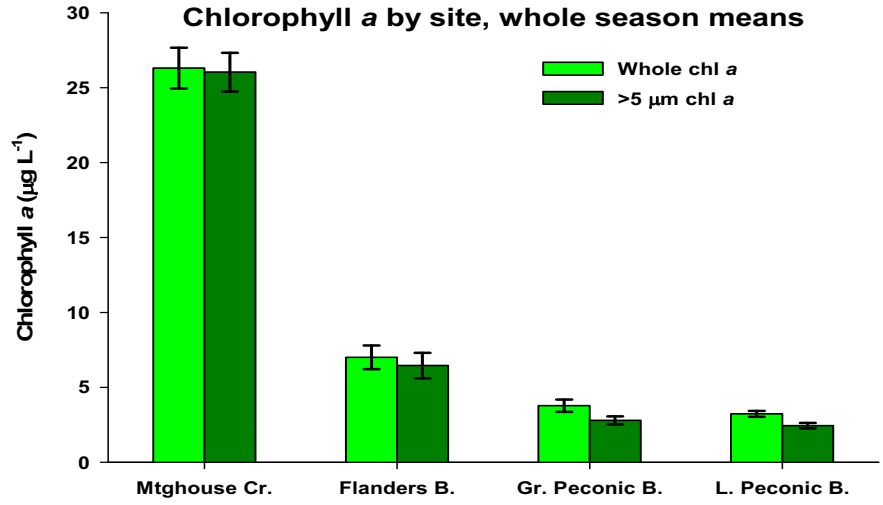

Bottom temperatures at field sites
Bottom temperature (°C)
28
26
24
22
20
18
16
14
12
10
8
Jun
Jul
Aug
Sep
Oct
Nov
Dec
Mtghouse Cr.
Flanders B.
Gr. Peconic B.
L. Peconic B.
Bottom salinities at field sites
Salinity (psu)
32
30
28
26
24
22
20
18
16
Jun
Jul
Aug
Sep
Oct
Nov
Dec
Mtghouse Cr.
Flanders B.
Gr. Peconic B.
L. Peconic B.
Dissolved oyxgen near bottom
Dissolved oxygen (mg L-1)
12
10
8
6
4
2
0
Jun
Jul
Aug
Sep
Oct
Nov
Dec
Date
Mtghouse Cr.
Flanders B.
Gr. Peconic B.
L. Peconic B.

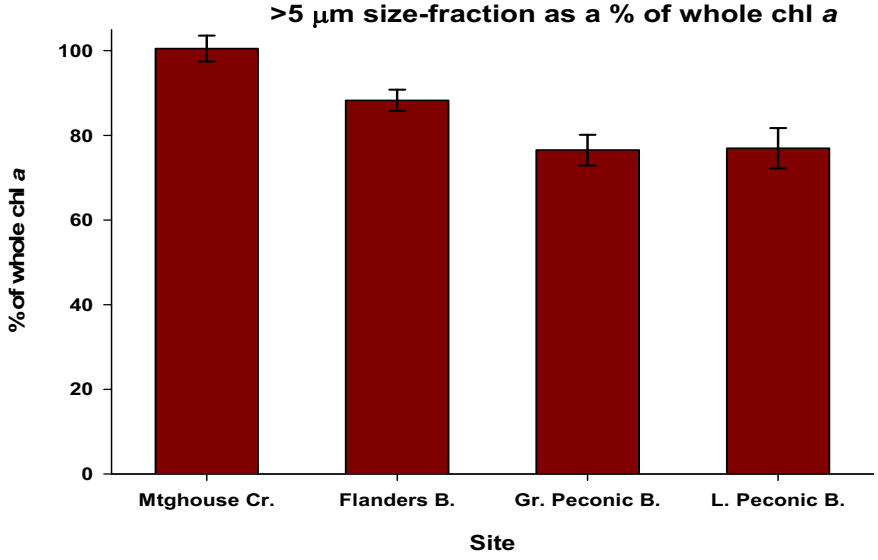
1

2

Figure 3 A-C. Temperature, salinity, and dissolved oxygen trends at 2 m depth, which is near the bottom at all field sites. The shellfish cage at Mtghouse Cr. was elevated to ~ 1 m, to avoid the hypoxia at 2 m.


Chlorophyll a time series
Chlorophyll a (µg L-1)
40
30
20
10
0
Jun
Jul
Aug
Sep
Oct
Nov
Dec
Date
Mtghouse Cr.
Flanders B.
Gr. Peconic B.
L. Peconic B.


Chlorophyll a by site, whole season means
Chlorophyll a (µg L-1)
30
25
20
15
10
5
0
Whole chl a
>5 µm chl a
Mtghouse Cr.
Flanders B.
Gr. Peconic B.
L. Peconic B.


>5 µm size-fraction as a % of whole chl a
% of whole chl a
100
80
60
40
20
0
Mtghouse Cr.
Flanders B.
Gr. Peconic B.
L. Peconic B.
Site

1

Figure 4 A-C. Chlorophyll *a* trends (mean ± SE) by site and by time series (A), averaged by site (B), and % in > 5µm size fraction (C).

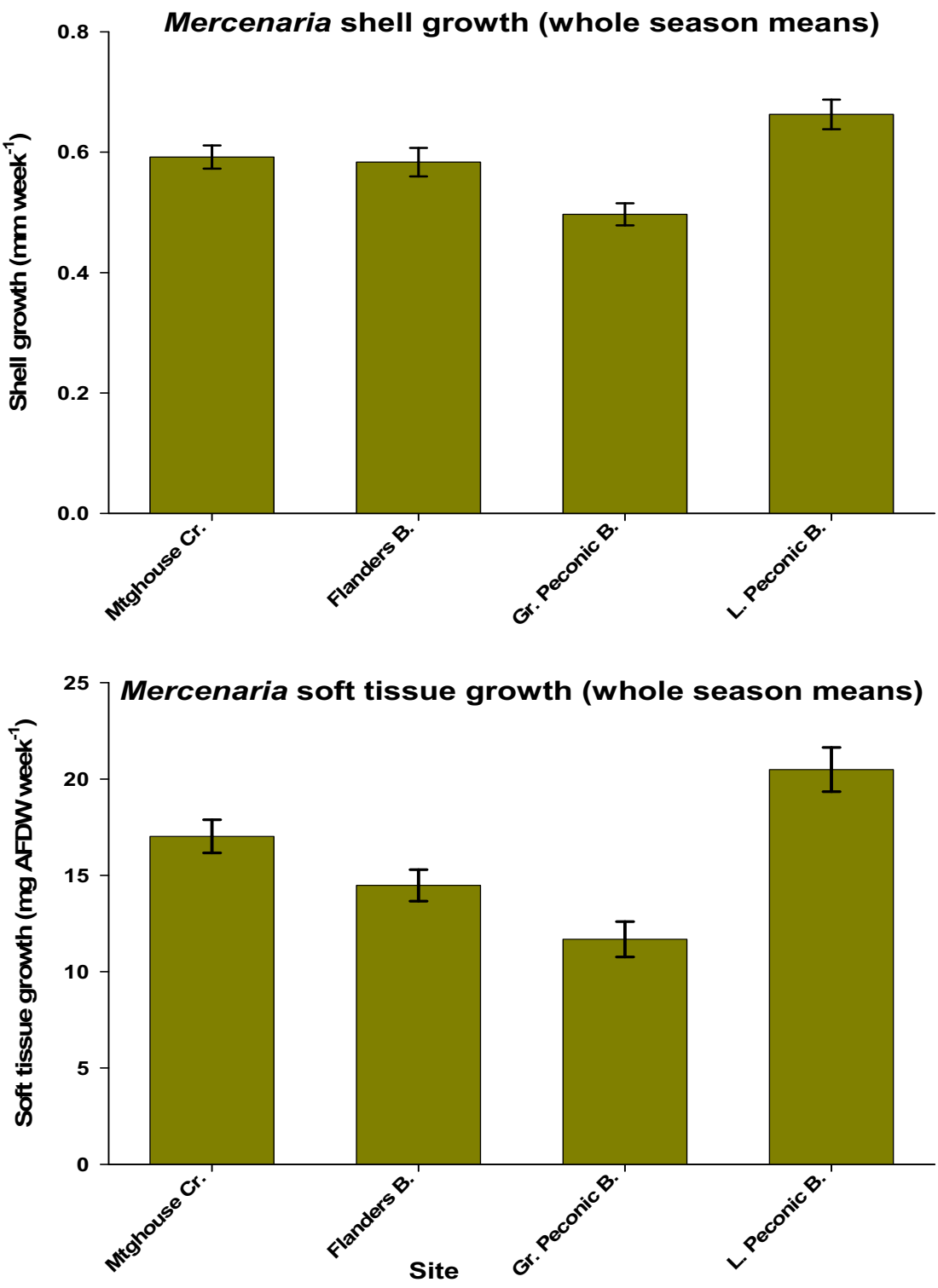


Figure 5 A-B. Juvenile *M. mercenaria* shell growth (A) and soft tissue growth (B) averaged over the whole season (Jun-Nov).

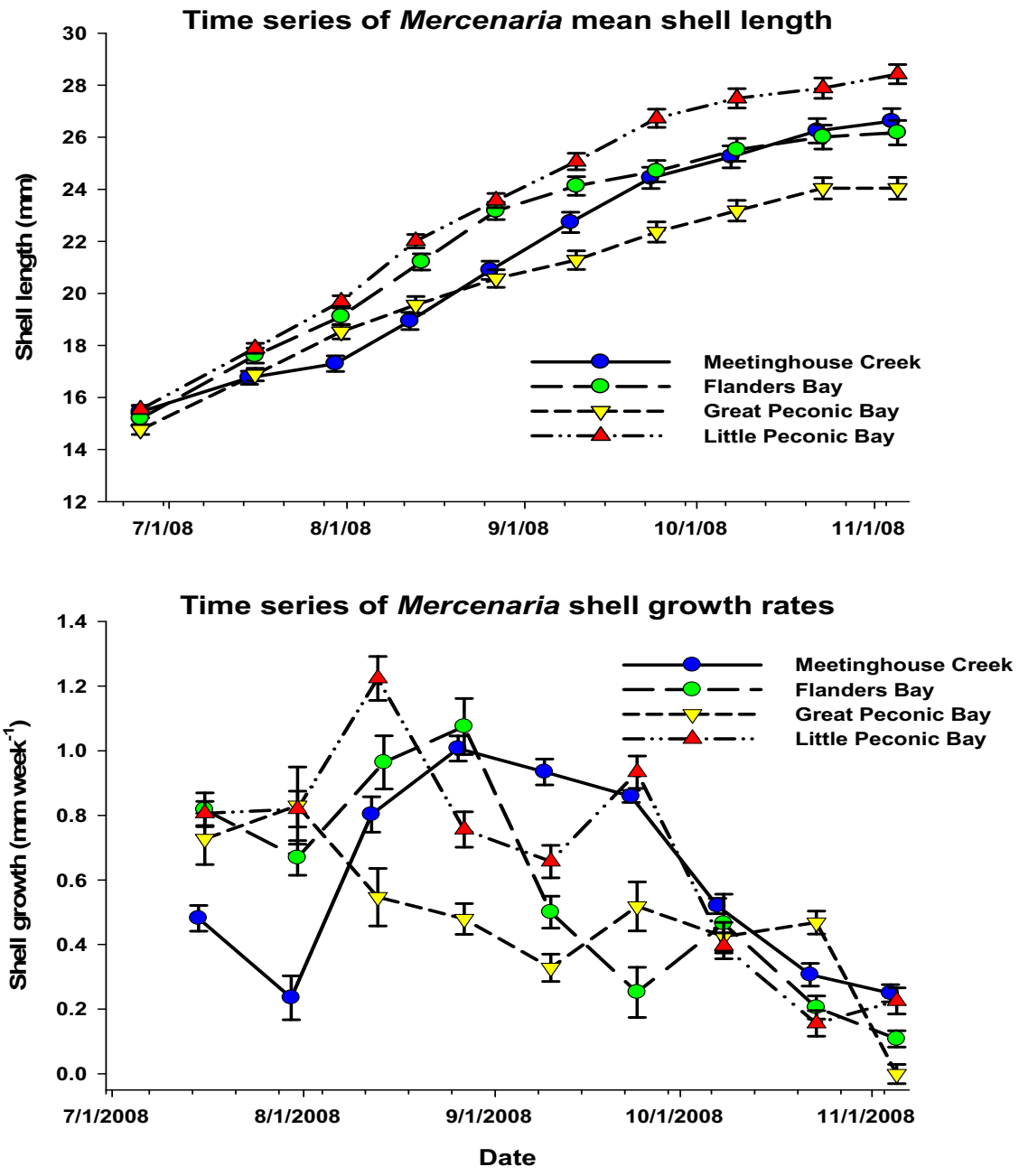


Figure 5 C-D. Juvenile *M. mercenaria* mean shell length (C) and shell growth (D) at each timepoint.

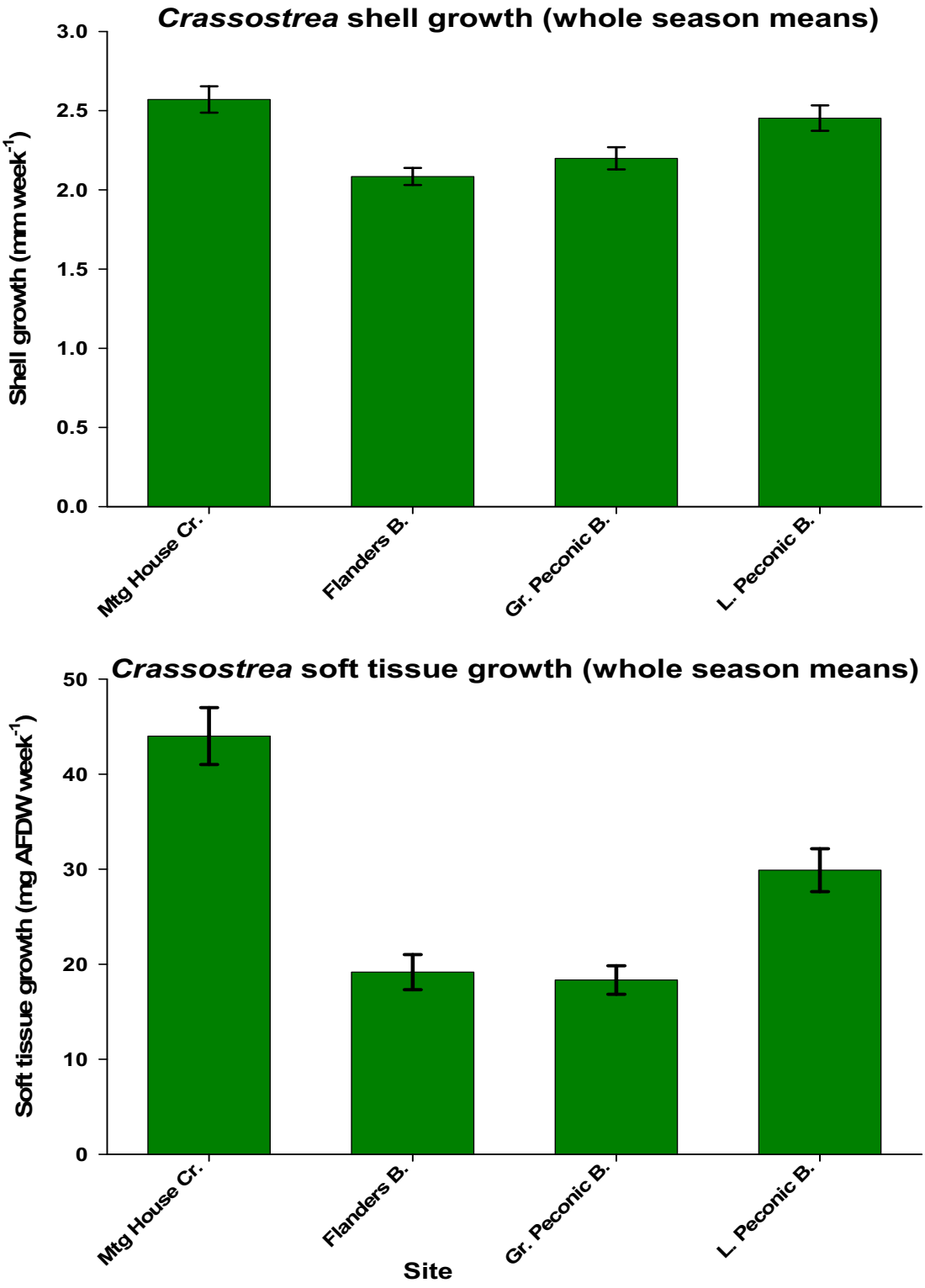


Figure 6 A-B. Juvenile *C. virginica* shell growth (A) and soft tissue growth (B) averaged over the whole season (Jun-Nov).

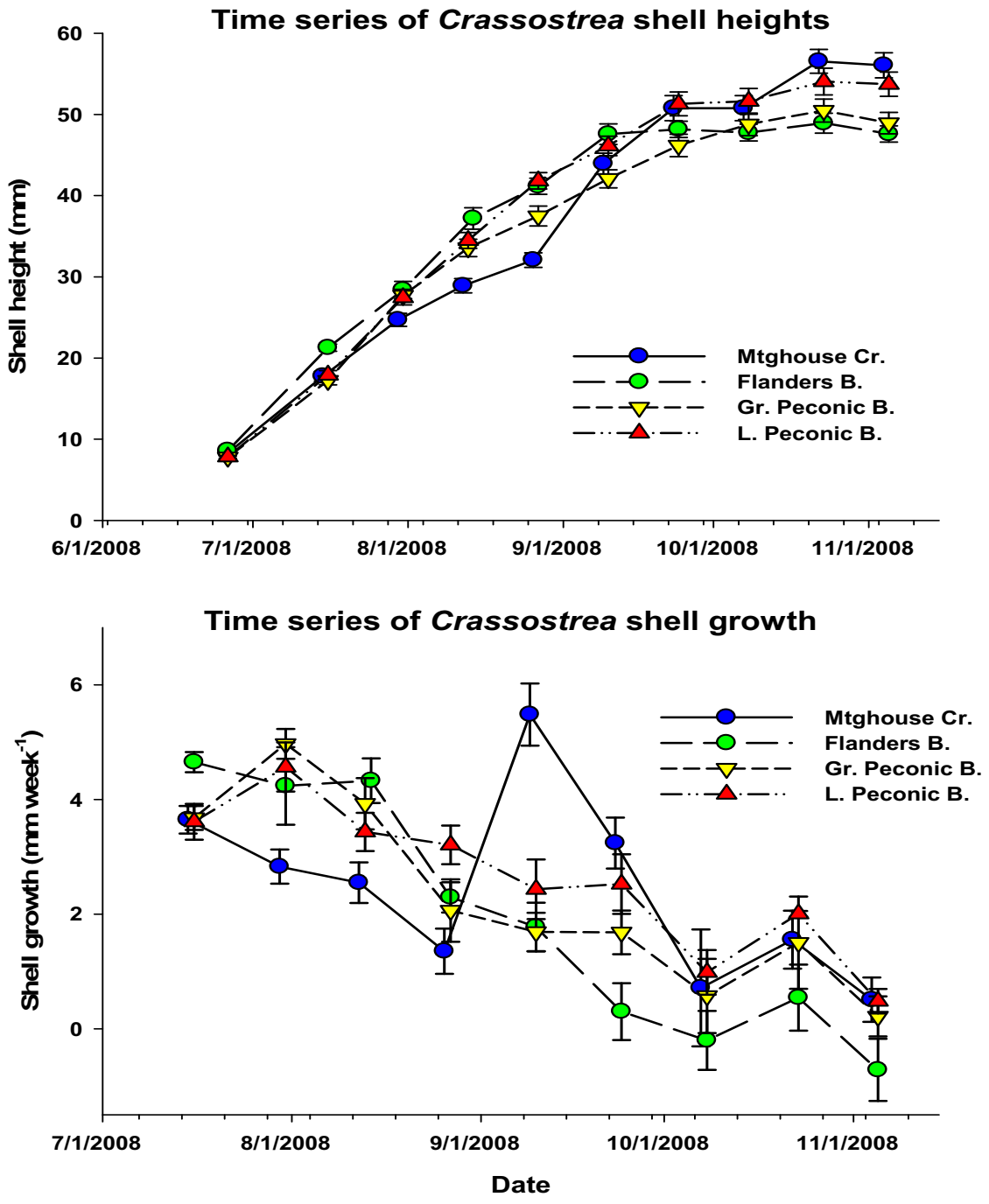


Figure 6 C-D. Juvenile *C. virginica* mean shell length (C) and shell growth (D) at each timepoint.

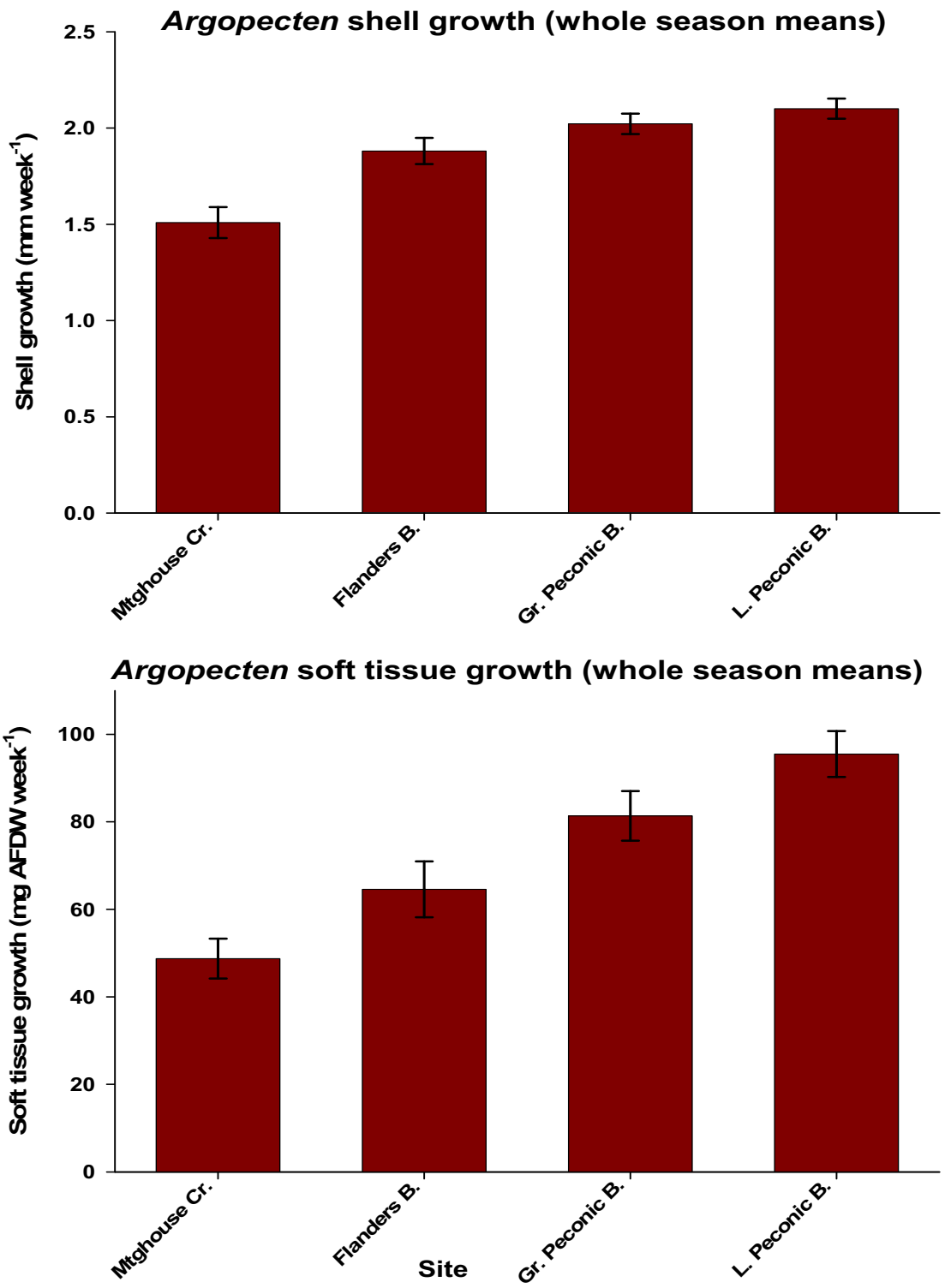


Figure 7 A-B. Juvenile *A. irradians* shell growth (A) and soft tissue growth (B) averaged over the whole season (Jun-Nov).

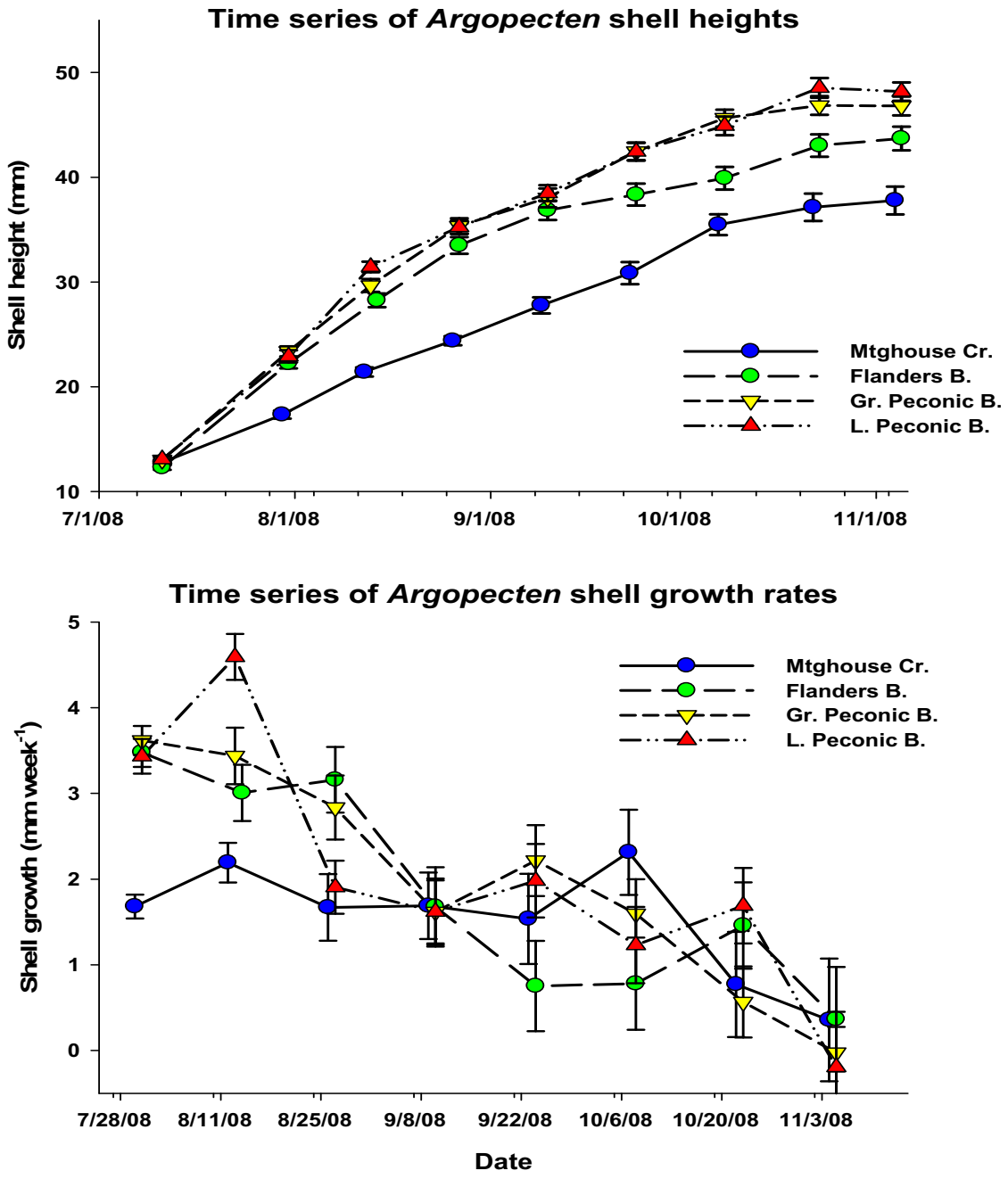


Figure 7 C-D. Juvenile *A. irradians* mean shell length (C) and shell growth (D) at each timepoint.

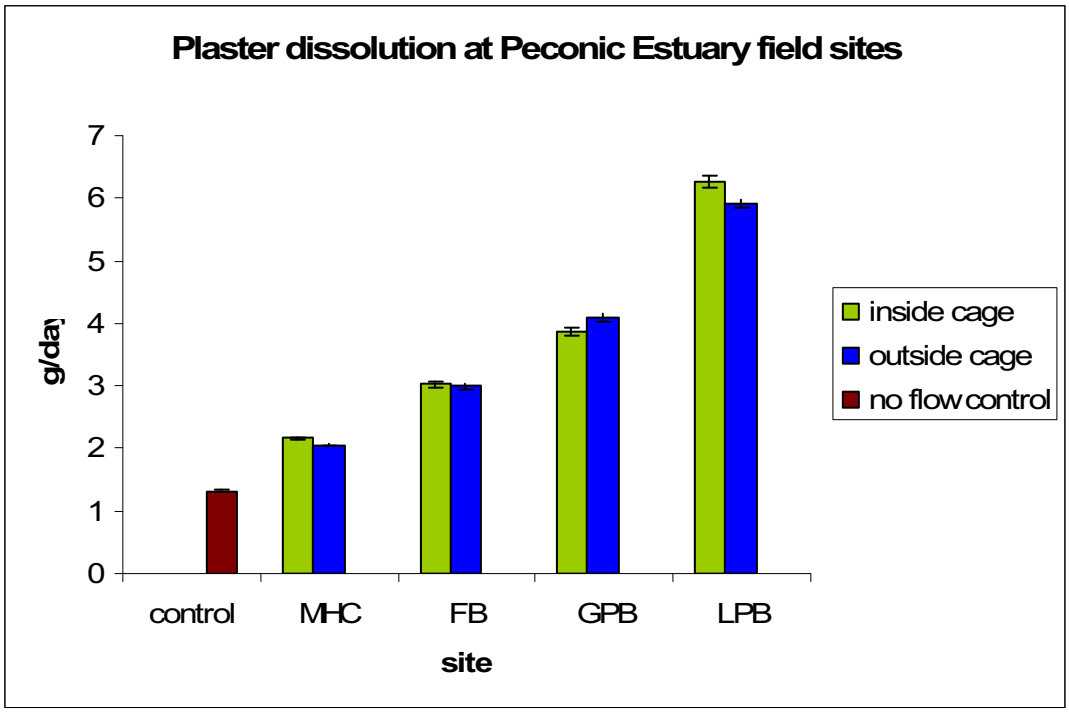
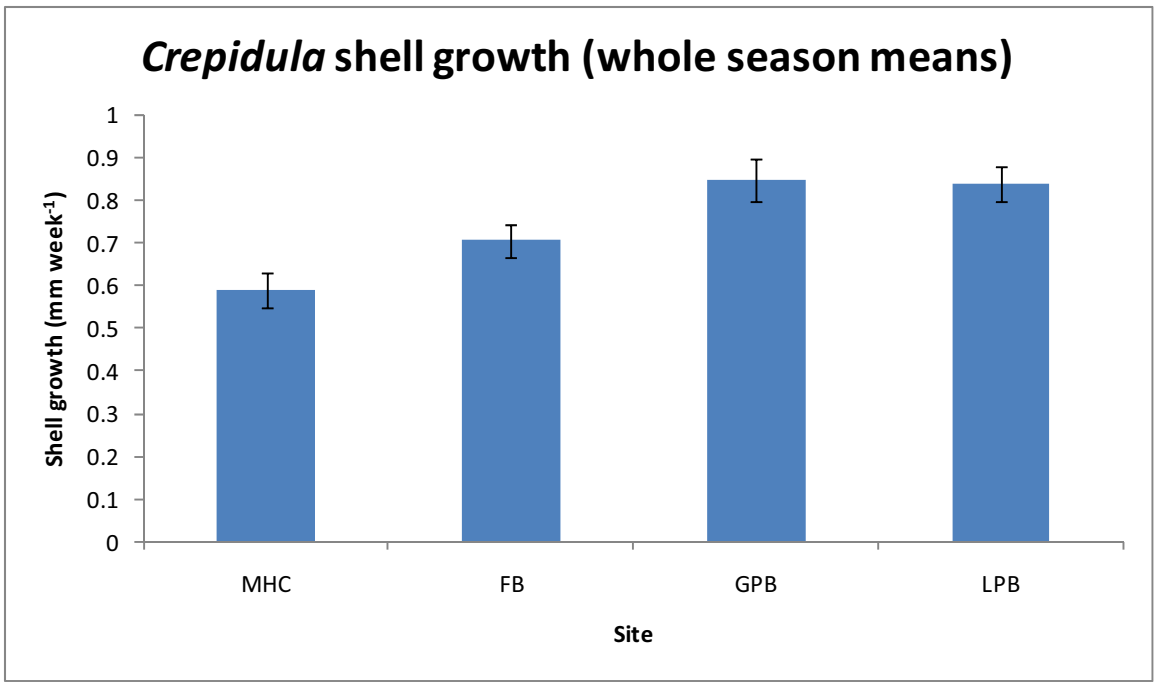


Figure 8. Dissolution of plaster blocks in g d^{-1} (mean ± SE) placed at field stations and in a "no flow" control tank. Dissolution measurements were averaged over 24 h from Oct 23rd-24th, 2008.



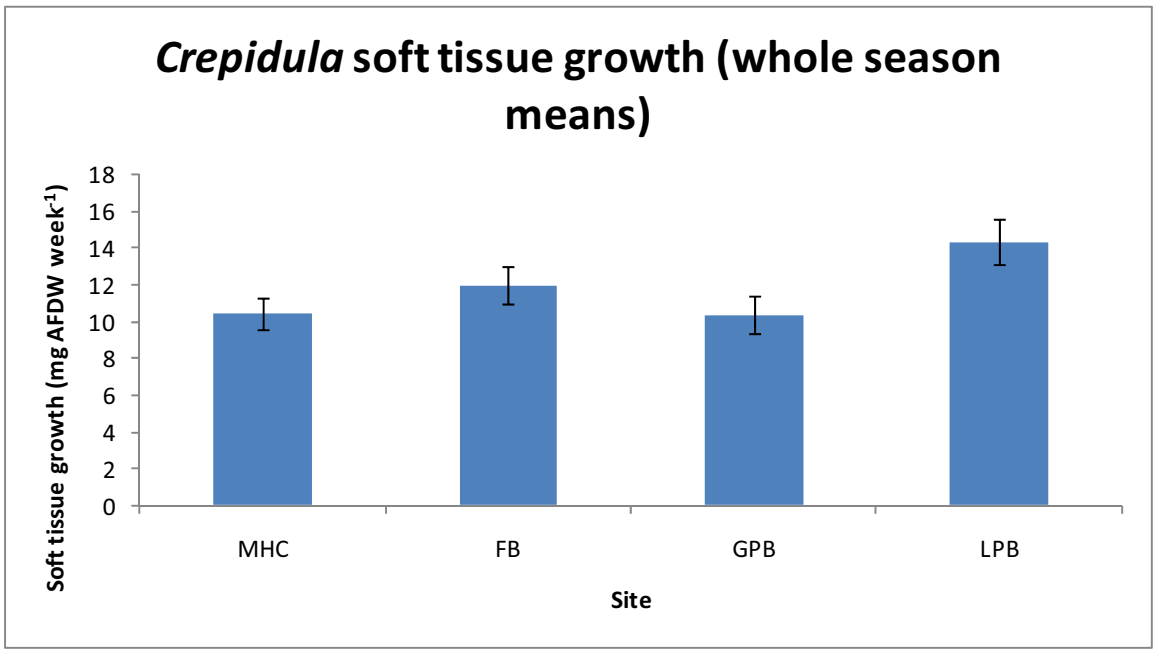
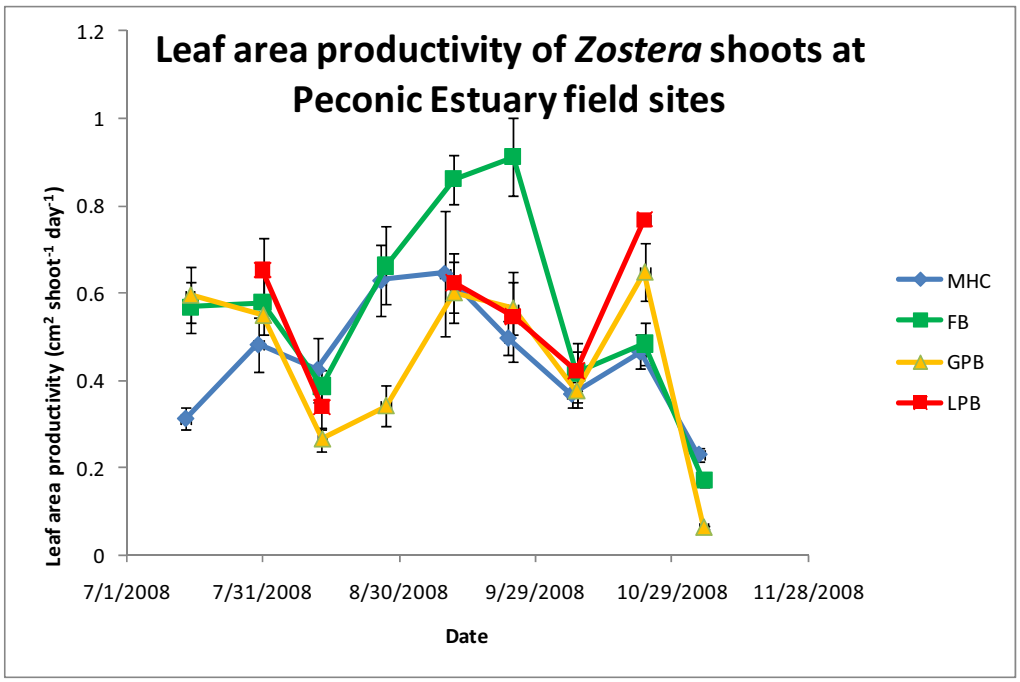


Figure 9 A-B. Juvenile *C. fornicata* shell growth (A) and soft tissue growth (B) averaged over the whole season (Jun-Nov).



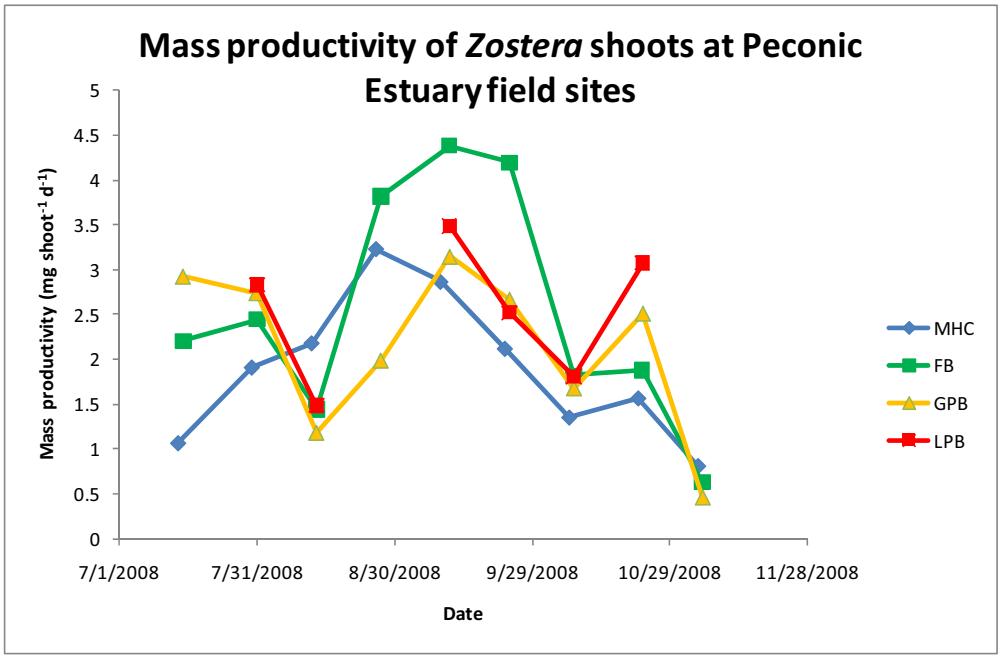


Figure 10 A-B. Time series of eelgrass (*Z. marina*) growth rates at Peconic Estuary field sites measured by leaf area productivity (A, means ± SE) and mass productivity (B, means).

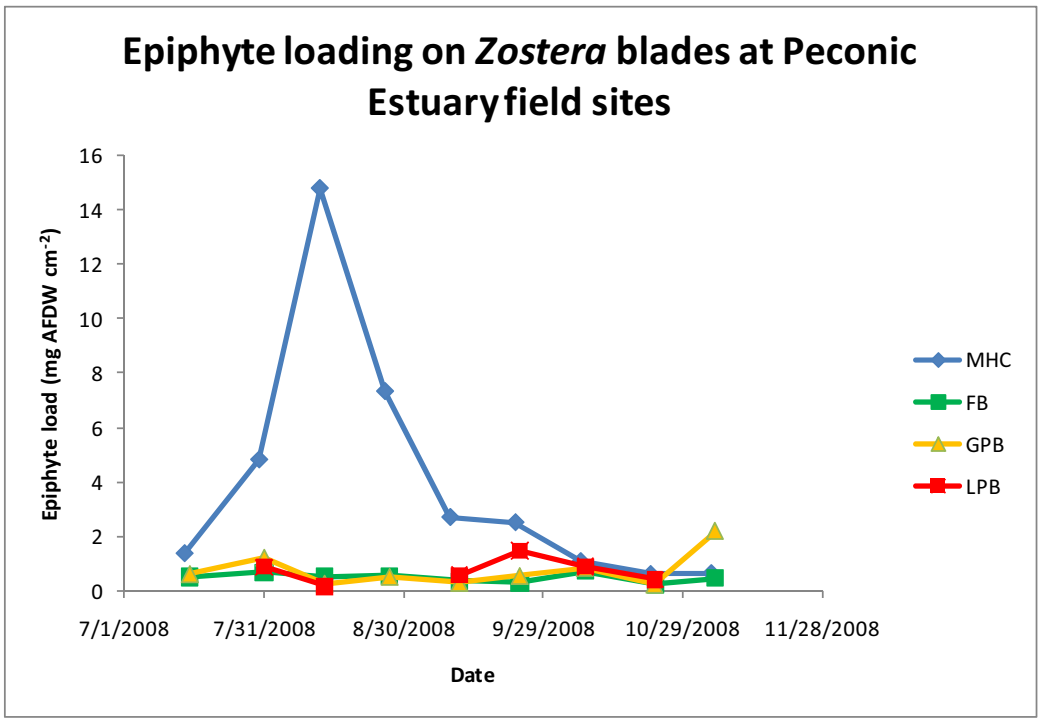


Figure 11. Time series of epiphyte densities on eelgrass (*Z. marina*) blades at Peconic Estuary field sites.

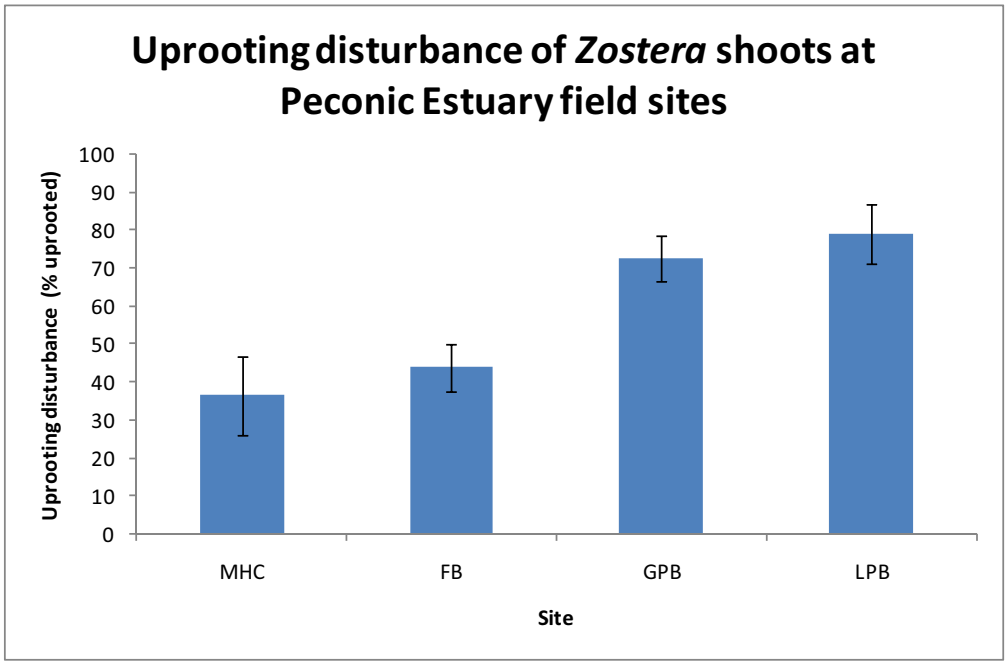


Figure 12. Uprooting disturbance of eelgrass shoots (*Z. marina*) at Peconic Estuary field sites. Values are means ± SE averaged over the whole season (Jun-Nov).

Chapter II:

The growth of estuarine resources (*Zostera marina*, *Mercenaria mercenaria*, *Crassostrea virginica*, *Argopecten irradians*, *Cyprinodon variegatus*) in response to nutrient loading and enhancement of suspension-feeding by adult shellfish

This is manuscript based on field work conducted in 2007 for the TNC project. It was submitted to Aquatic Biology in April 2008.

Keywords: *Zostera marina, Crassostrea virginica, Mercenaria mercenaria, Argopecten irradians, Cyprinodon variegatus,* eelgrass, seagrass, clams, oysters, eutrophication, nutrients, nutrient-loading, aquaculture, bivalves, suspension feeders, mesocosms, ecosystem-based management, estuarine restoration

Abstract: While many coastal ecosystems previously supported high densities of seagrass and resource bivalves, the impacts of overfishing, eutrophication, harmful algal blooms, and habitat loss have contributed to the decline of these important resources. Despite improvements in wastewater treatment in some watersheds and subsequent reduced nutrient loading to neighboring estuaries, seagrass and bivalve populations in these locations have generally not recovered. We conducted a series of mesocosm experiments to simultaneously examine the effects of nutrient loading and historic suspension-feeder densities on the growth of eelgrass (*Zostera marina*), juvenile bivalves (hard clams, *Mercenaria mercenaria*, Eastern oysters, *Crassostrea virginica*, and bay scallops, *Argopecten irradians*), and juvenile fish (sheepshead minnow, *Cyprinodon variegatus*). High nutrient loading rates led to significantly higher chlorophyll *a* levels and significantly increased growth of juvenile bivalves relative to controls with lower nutrient loading rates. The filtration provided by adult suspension-feeders (*M. mercenaria* and *C. virginica*) significantly decreased chlorophyll *a* levels, significantly increased light penetration and the growth of eelgrass, and significantly decreased the growth of juvenile bivalves and fish relative to controls with no filtration from adult suspension-feeders. These results suggest that either nutrient loading or bivalve filtration or both have the capacity to structure estuarine food webs and affect the recovery of estuarine resources. Future ecosystem-based approaches will need to account for both anthropogenic nutrient-loading and bivalve restoration to successfully manage estuarine resources.

Introduction:

Estuaries are home to a variety of valuable living resources. Finfish and shellfish are harvested directly in commercial and recreational fisheries, while seagrass beds are considered of paramount importance as structural habitat for shellfish and finfish in many coastal areas (Heck and Wetstone 1977, Irlandi and Peterson 1991, Beck *et al.* 2001). Many of the world's estuaries currently support lower abundances of finfish, shellfish, and seagrasses than they did historically due to overfishing (Jackson *et al.* 2001, Lotze *et al.* 2006), habitat loss (Orth *et al.* 2006), eutrophication (Nixon 1995, de Jonge *et al.* 2002), and harmful algal blooms (Hallegraeff 1993, Gobler *et al.* 2005, Sunda *et al.* 2006). As such, estuarine management plans are typically focused on combating these harmful processes and restoring living resources (Cloern 2001, Newell 2004, Lotze *et al.* 2006).

Changes in inorganic nutrient loading to estuaries can indirectly influence the growth of marine resource species. High rates of nutrient loading have been associated with increases in pelagic productivity, decreased water clarity, hypoxia, and declines in seagrass growth and abundances (Duarte 1995, Diaz and Rosenberg 2008, Wall *et al.* 2008). In response, estuarine management efforts often focus primarily on reducing anthropogenic nutrient loading in an effort to curb the negative effects of eutrophication (Cloern 2001, de Jonge *et al.* 2002). However, some level of nutrient loading must be necessary to sustain primary and secondary production (Nixon and Buckley 2002). High levels of inorganic nutrients favor larger phytoplankton cells (Malone 1980, Raven and Kubler 2002), such as diatoms and prymnesiophytes, which are generally considered a good source of nutrition for bivalves (Beukema and Cadee 1991, Wikfors *et al.* 1992, Weiss *et al.* 2007). Studies in several estuaries have shown blue mussels (*Mytilus edulis*), hard clams (*Mercenaria mercenaria*), and softshell clams (*Mya arenaria*) can respond

positively to increased nitrogen loading and high chlorophyll *a* levels in their habitats (van Stralen and Dijkema 1994, Weiss *et al.* 2002, Carmichael *et al.* 2004, Weiss *et al.* 2007). Weiss *et al.* (2002) and Carmichael *et al.* (2004) found that shell growth, soft tissue growth, and survival of *M. mercenaria* and *M. arenaria* increased along a naturally-occurring gradient of nitrogen loading in Waquoit Bay, Massachusetts, USA. They attribute these changes to increased quantity and quality of food particles due to nitrogen enrichment (Carmichael and Valiela 2005), although a similar response has not been found for bay scallops (*Argopecten irradians*, Shriver *et al.* 2002). While nutrient over-loading in estuaries has a well-known set of negative consequences (Valiela *et al.* 1992, Nixon 1995, Kemp *et al.* 2005), the stimulation of secondary production in bivalves could be an overlooked positive effect of nutrient loading (Nixon and Buckley 2002, Carmichael *et al.* 2004, Carmichael and Valiela 2005), especially in shallow ecosystems with well-mixed water columns that rarely experience hypoxia.

As described in many studies and reviews, suspension feeding bivalves can have a variety of positive effects on estuaries, such as reducing phytoplankton biomass (Officer *et al.* 1982, Doering and Oviatt 1986), transferring nutrients and biomass to the benthos (Smaal and Prins 1993, Jackson 2001), control of harmful algae (Cerrato *et al.* 2004), increased light penetration (Newell and Koch 2004), and facilitating the growth of benthic plants (Peterson and Heck 2001, Newell *et al.* 2002, Wall *et al.* 2008). As bivalve populations have declined through overfishing, habitat loss, and disease, these ecosystem services have been lost from estuaries (Newell 1988, Lotze *et al.* 2006). In the absence of dense natural bivalve populations, bivalve aquaculture may be a tool for managers to restore ecosystem functions previously provided by natural populations (Newell 2004, Ruesink *et al.* 2005), combat eutrophication (Gifford *et al.* 2004, Cerco and Seitzinger 2007), or ease harvest pressures on wild populations (Dolmer and Frandsen 2002).

Aquaculture is on the rise world-wide, and bivalve aquaculture may avoid some of the pitfalls of finfish aquaculture (Naylor *et al.* 2000) while controlling phytoplankton blooms and affecting carbon and nutrient cycling in ways that are comparable to natural shellfish populations (Newell 2004, Zhou *et al.* 2006, Huang *et al.* 2008).

Commercial bivalve aquaculture operations strive to grow a maximum number of shellfish in a minimum of space (Frechette *et al.* 1992), with locally high filtration rates sometimes leading to "self-thinning" through density-dependent food limitation (Rheault and Rice 1996, Zhou *et al.* 2006). It is not well-known how these locally high filtration rates interact with adjacent natural bivalve populations (Ferreira *et al.* 2008), but locally high biodeposition rates from aquaculture have produced negative effects in some systems (Newell 2004 and references therein). As aquaculture develops for both commercial and restoration purposes, an improved understanding of these effects will help managers use bivalves to achieve healthy ecosystem functions.

This study was designed to examine the combined effects of nutrient loading and adult bivalve filtration on the growth and survival of estuarine resource species: juvenile hard clams (*M. mercenaria*), bay scallops (*A. irradians*), and oysters (*C. virginica*), a juvenile planktivorous fish (sheepshead minnow, *Cyprinodon variegatus*), and eelgrass (*Z. marina*). These five species were placed into an array of mesocosms with treatments of high or low nutrient loading and presence or absence of adult bivalves arranged in a 2 x 2 factorial design. The growth of all populations along with levels of light, nutrients, size-fractionated chlorophyll *a*, and particulate organic matter were monitored during three experiments which demonstrated that both nutrient loading and adult bivalve filtration can strongly influence the growth of multiple estuarine resources.

Methods:

We conducted three mesocosm experiments placed within eastern Shinnecock Bay at the Stony Brook-Southampton Marine Science Center from June 5th, 2007, to September 6th, 2007. Shinnecock Bay is part of Long Island's south shore estuary lagoons which have followed a trajectory in the decline of resources common to many estuaries around the world (Bricelj and Kuenster 1989, McHugh 1991, Gobler *et al.* 2005). Specifically, these lagoons have seen declines in shellfish such as the hard clam (*Mercenaria mercenaria*, McHugh 1991), the bay scallop (*Argopecten irradians*, Bricelj and Kuenster 1989), various finfish (Beck *et al.* 2001), and eelgrass beds (*Zostera marina*, Dennison *et al.* 1989). The 300-L mesocoms used in this study have been utilized previously to yield realistic growth rates and conditions for seagrass and shellfish (Cerrato *et al.* 2004, Wall *et al.* 2008). The depth of the mesocosms (1.2 m) is within the range of the mean depths found among Long Island's south shore estuary lagoons (Wilson *et al.* 1991). Moreover, the placement of the tanks in eastern Shinnecock Bay allowed for ambient light and temperature to be maintained during experiments. Replicate experimental mesocosms (n = 4 for each treatment) were stocked with juvenile hard clams (~10 mm), scallops (~10 mm), and/or oysters (~10 mm) at stocking densities (10 – 20 $tank^{-1}$ or 36 – 72 m^{-2}; Table 1) more than an order of magnitude lower than standard commercial aquaculture stocking densities (~500 individ. m^{-2}; Barber and Davis 1997, Kraeuter and Castagna 2001) to avoid inter- and intraspecific competition for food (Rheault and Rice 1996, Kraeuter and Castagna 2001). Indeed, our estimated community clearance rates of juvenile bivalves indicated they filtered 0.4-1.5% d^{-1} of the total mesocosm volumes. All juvenile bivalves were placed in mesh cages (~2mm mesh size) near the bottom of the mesocosms. Juvenile shellfish were obtained from the

Cornell Cooperative Extension shellfish hatchery in Southold, NY. Three-week-old sheepshead minnows (*Cyprinodon variegatus,* 10-15 mm) were obtained from Cosper Environmental Services in Bohemia, NY. These planktivorous fish (Samson *et al.* 2008) were held in mesh baskets suspended near the tops of the experimental tanks (n = 10). A laminar circulating pump (Rio 180) was utilized to ensure mesocosms were well-mixed. In addition to the suspension feeders, individual shoots of *Zostera marina* (n = 16) were transplanted into planters containing low-organic sand and placed in each mesocosm (Wall *et al.* 2008).

Mesocosms were filled with eastern Shinnecock Bay water during high tide. Water from this region is fairly mesotrophic with mean total N concentrations of 0.2 ± 0.1 mg N L^{-1} or 16 ± 8 µM N from 2000 to 2005 (n = 50 measurements; SCDHS 2000-2005). Actual measurements of each inorganic nitrogen, phosphorus and silicon species were made at the beginning of each experiment, in the middle of experiments, and on the final day using standard techniques (Valderrama 1981, Parsons *et al.* 1984). For each experiment, we established a low nutrient loading rate for half of the experimental tanks (0.065-0.255 mmoles N m^{-2} d^{-1}) using a 1-2% d^{-1} exchange with Shinnecock Bay water. The other half of the tanks received a high nutrient loading rate (5.49-10.70 mmoles N m^{-2} d^{-1}) that reflected ambient exchange plus nutrient additions of ammonium and the Redfieldian equivalent of orthophosphate. These nutrient loading rates are within the range found in more eutrophic Northeast US estuaries such as the Childs River, MA, and Moriches Bay, NY (Taylor *et al.* 1999). Nutrient stocks were filter-sterilized (0.2 µm) and stored frozen. Experiments were run in semi-continuous mode, with 1-2% of the water volume being replaced daily mimicking the natural tidal exchange which occurs in the back-bay regions of the Peconic Estuary and Great South Bay (Hardy 1976, Wilson *et al.* 1991). For each experiment, half of the experimental tanks contained adult suspension feeders

(hard clam *M. mercenaria* or Eastern oyster *C. virginica*) and half of the tanks contained no adult suspension feeders. Adult hard clams measured 56.70 ± 1.18 mm anterior-posterior and weighed 1.64 ± 0.11 g AFDW. Adult oysters measured 59.17 ± 0.79 mm hinge-ventral margin and weighed 0.66 ± 0.05 g AFDW. Adult shellfish were locally caught and obtained from seafood markets. The filtration of adult shellfish was estimated with a clearance rate method (Riisgard 2001) using water (>15 μg L^{-1} chlorophyll *a*) from the experimental tanks. Clearance rates were calculated according to the equation:

clearance rate = ln(initial chl *a* / final chl *a*) * volume / time

This measurement was performed once per species. A 'community' clearance rate was estimated from these data using the average individual clearance rate and the number of individuals in the tank. An estimated turnover time for the entire tank volume to pass through the adult shellfish was calculated for each tank by dividing the tank volume by this community clearance rate. A summary of experimental conditions for all three experiments is presented in Table 1.

Experiments were conducted for ~two weeks, and shellfish growth was assessed via the changes in ash-free dry weight (AFDW) of tissue or by changes in lengths between initial and final individuals within each mesocosm (Weiss *et al.* 2007). The length of juvenile clams was measured by shell length (anterior-posterior; Kraeuter and Castagna 2001) and the size of juvenile oysters and scallops was measured by shell height (hinge-ventral margin; Rheault and Rice 1996). Bivalve tissue was dried at 70º C for at least 24 h and then ashed at 450º C for an additional 4 h (Gabbott and Walker 1971, Bass *et al.* 1990). One hundred bivalves of each species were selected from the initial set to provide a mean initial AFDW. When fewer than 100 individuals were available for a mean initial AFDW, initial AFDW's were hind-casted based on initial lengths using length-weight regressions from 100+ individuals of the same species and

size class. Juvenile fish growth was measured by total length only. Mean growth rates for all species based on length or weight were calculated by the change in length or AFDW divided by the number of days between initial and final measurements. In addition, treatment effects on eelgrass above-ground biomass, leaf morphometrics, leaf productivity and epiphyte biomass were assessed by marking then harvesting eelgrass shoots from each replicate mesocosm. Leaf production during the experiment was measured using a modified leaf marking technique (Ibarra-Obando and Boudouresque 1994). Sixteen eelgrass shoots were marked at the base of the leaves by driving an 18-gauge hypodermic needle through all of the leaves on the shoot. The marked shoots were allowed to grow for 14 d, after which all above-ground leaf material was harvested. In the laboratory, the number of leaves, leaf width, leaf length, daily gross above-ground production, and leaf epibiont biomass (mg AFDW cm^{-2} leaf area) was determined. Productivity was determined by both mass (mg $shoot^{-1}$ d^{-1}) and leaf area growth (cm^2 $shoot^{-1}$ d^{-1}). Epiphyte mass was determined by scraping fouling organisms and algae from each leaf, then drying them to a constant mass (± 0.01 mg) in an oven at 70 ^{o}C. Following dry weighing, the ash weight was determined by ashing the samples at 450 ^{o}C for 4 h. Ash free dry weight (AFDW) was calculated as dry weight minus ash weight.

Bottom mesocosm light levels were measured every 15 minutes by HOBO© Pendant-style data loggers with light sensors. A data logger was placed in each experimental tank near the bottom at a depth of approximately 1 m, a height just above eelgrass and shellfish cages preventing the obstruction of incoming light. A mean daily light level for each experimental tank was calculated by averaging values between 1000 and 1400 hrs, when the sun was most directly overhead. Since the HOBO© data loggers measure visible light levels in Lux instead of photosynthetically active radiation (PAR) in μmol m^{-2} s^{-1}, we compared measurement of light

with the HOBO© loggers to those obtained with a LiCor© LI-192 underwater quantum sensor of PAR. There was a highly significant linear relationship between visible light in Lux as measured by the HOBO© data logger and PAR as measured by the LiCor© sensor over depths of 0.5-2.0 m (Visible light in LUX = 41.407 * PAR – 408.67, $r^2 = 0.98$, $p < 0.001$). Based on this finding, we believe that experimental light readings from HOBO© data loggers within our mesocosms were representative of the general trends in PAR.

The quality and quantity of suspended food for bivalves was assessed in two ways. The distribution of phytoplankton biomass among size classes was estimated by the analysis of size fractionated chlorophyll *a* (whole and >5 μm) using polycarbonate filters and standard fluorometric techniques (Parsons *et al.* 1984). Whole water samples were collected on pre-combusted glass fiber filters for the analysis of particulate organic carbon and nitrogen (POC, PON) on a CE Instruments Flash 1112 elemental analyzer (Sharp 1974, Gobler and Sañudo-Wilhelmy 2001).

Seawater dilution experiments were conducted to quantify the rates of microzooplankton grazing of algal biomass within the mesocosm tanks (Landry *et al.* 1995). During each experiment, five liters of water from each replicate mesocosm within a treatment were pooled into a 20 L carboy for that treatment. Triplicate samples of 100, 70, 40 and 15% experimental dilutions of whole seawater with filtered seawater (0.2 μm) from each carboy were established in 1 L polycarbonate bottles. To ensure nutrient-replete growth during these experiments, nitrate (20μM) and orthophosphate (1.25μM) was added to all of the bottles. A set of triplicate controls of whole seawater without nutrients were also established for each grazing experiments (Landry *et al.* 1995). Algal growth rates (μ) within experimental bottles were quantified using the formula: $\mu = [\ln(B_t / B_o)] / t$, where μ is the net growth rate, B_t is the amount of biomass (chl *a*)

present at the end of the experiments, B_o represents the amount of biomass at the beginning of experiments, and t is the duration of the experiment in days. The slope of first order linear regressions of dilution of seawater (x-axis) and the net growth rates (y-axis) were used to establish grazing mortality rates (Landry *et al.* 1995).

Statistical Analysis

Differences in the growth of each animal species and eelgrass was assessed by means of Two-Way ANOVAs, with nutrient loading level and presence/absence of adult bivalves as the two treatment factors. Chlorophyll *a* and light level trends were analyzed using three-way ANOVAs with nutrient loading, presence/absence of adult bivalves, and day as factors. When a significant effect on the response variables was detected, multiple comparison tests (Tukey's Studentized range) was used to test for significant differences between levels within the treatment. Mortality of juvenile bivalves was analyzed using a G-test of independence (Sokal and Rohlf 1995). All statistical results were considered significant if $p < 0.05$.

Results:

Experiment 1

Three separate mesocosm experiments were carried out using the above methods (Table 1). Experiment 1 ran from June 5th to June 18th, 2007. The average temperature in the experimental tanks was 20.30 ± 0.15 °C, the average salinity was 26.42 ± 0.04, and the average dissolved oxygen was 6.65 ± 0.14 mg L^{-1}. The "low nutrient loading" treatment received an average of 0.065 mmoles N m^{-2} d^{-1} through a 1% d^{-1} exchange with Shinnecock Bay water whereas the "high nutrient loading" treatment received 10.70 mmoles N m^{-2} d^{-1}. The densities of adult suspension feeders were 29 or 0 clams m^{-2} (*M. mercenaria*). The estimated turnover time

from bivalve filtration for the experimental tanks with hard clams was 2.4 days. All tanks in this experiment were stocked with juvenile clams, juvenile oysters, and eelgrass (Table 1).

In this experiment, the higher nutrient loading rate (10.70 mmoles N m^{-2} d^{-1}) and the absence of adult hard clams produced significant increases in chlorophyll *a* compared to the low nutrient loading rate (0.065 mmoles N m^{-2} d^{-1}) and the presence of adult hard clams (29 individ. m^{-2}) over the course of a 13-day experiment (Fig. 1; $p<0.001$, 3-way ANOVA). Chlorophyll *a* in the <5 μm size fraction was significantly increased by high nutrient loading ($p<0.01$) and decreased by adult clam filtration ($p<0.001$, 3-way ANOVA), while the >5 μm size fraction was not significantly affected by the experimental treatments (3-way ANOVA). Particulate organic nitrogen (PON) was significantly higher in the presence of adult clams (25.17 ± 2.57 μM) compared to the absence of adult clams (17.28 ± 1.70 μM; $p<0.05$, 2-way ANOVA). Although particulate organic carbon (POC) did not change between treatments, the molar ratio of POC:PON was significantly higher under low nutrient loading (9.70 ± 0.61) and the absence of adult clams (10.51 ± 0.45) compared to high nutrient loading (9.22 ± 0.26) and the presence of adult clams (8.83 ± 0.24; $p<0.05$ for nutrient treatment, $p<0.01$ for clam filtration treatment, 2-way ANOVA). Mean chlorophyll *a* and POC/PON levels for all treatments are summarized in Table 3.

The highest juvenile hard clam growth was in the presence of high nutrient loading and in the absence of adult hard clams, while the lowest was without nutrient loading but with adult hard clams present (Fig. 2A) However, only the nutrient loading treatment had a statistically significant effect: juvenile clam growth (Fig. 2A) and juvenile oyster growth (Fig. 3B) were both significantly higher in the high nutrient loading treatment (0.032 ± 0.009 mm d^{-1} and 0.078 ± 0.016 mg AFDW d^{-1}, respectively) compared with treatments without experimental nutrient

addition (0.00 ± 0.01 mm d^{-1} and 0.034 ± 0.015 mg AFDW d^{-1} respectively; $p<0.05$ for each, 2-way ANOVA). Juvenile bivalve growth for both species was marginally lower when adult clams were present, but not significantly so (Fig. 2A, 2B). Despite the changes in chlorophyll *a*, light levels were not significantly different among treatments (3-way ANOVA) and subsequently eelgrass growth was not affected by the experimental treatments (2-way ANOVA). Nitrate, ammonium, and phosphate accumulated within the nutrient enriched mesocosms and to a lesser extent in the adult clam only treatment during this experiment, while silicate levels were similar among treatments (Table 2). Microzooplankton grazing rate data were not available for this experiment.

Experiment 2

Experiment 2 ran from July 12th to July 27th, 2007. The average temperature in the experimental tanks was 24.27 ± 0.16 °C, the average salinity was 28.02 ± 0.16, and the average dissolved oxygen was 5.83 ± 0.12 mg L^{-1}. The "low nutrient loading" treatment received an average of 0.255 mmoles N m^{-2} d^{-1} through a 2% d^{-1} exchange with Shinnecock Bay water. The "high nutrient loading" treatment received ambient exchange plus a daily experimental nutrient addition for a total of 5.75 mmoles N m^{-2} d^{-1}. The densities of adult suspension feeders were 21 or 0 oysters m^{-2} (*C. virginica*). The estimated turnover time from bivalve filtration for the experimental tanks with oysters was 1.5 days. All tanks in this experiment were stocked with juvenile clams, juvenile oysters, and eelgrass (Table 1).

Nitrate, phosphate, and silicate levels all increased during experiments, but did not differ among treatments, while ammonium levels were lowest in the no oyster, no nutrient treatment (Table 2). Treatments did not significantly affect total chl *a* levels or <5 µm chl *a* (3-way

ANOVA), but did affect >5 μm chl *a*, which was higher in the absence of adult oysters ($p<0.05$, 3-way ANOVA, Table 3). Juvenile clam growth was significantly higher in the high nutrient loading treatment (0.039 ± 0.003 mm d^{-1} and 0.058 ± 0.005 mg AFDW d^{-1}) compared to the low nutrient loading treatment (0.030 ± 0.003 mm d^{-1} and 0.033 ± 0.005 mg AFDW d^{-1}) when measured by shell length (data not shown; $p<0.05$, 2-way ANOVA) or by dry tissue weight (Fig. 3A; $p<0.001$, 2-way ANOVA). Juvenile clam growth was not affected by the adult oyster filtration treatment. In contrast, the juvenile oysters responded to the adult bivalve treatment; juvenile oyster growth was significantly decreased in the presence of adult oyster filtration (Fig. 3B; $p<0.01$; 2-way ANOVA) but was not affected by the nutrient loading treatments. Juvenile oyster growth was 0.131 ± 0.022 mg AFDW d^{-1} in the absence of adult oysters and was 0.033 ± 0.017 mg AFDW d^{-1} in the presence of adult oysters. Light levels and eelgrass growth were not significantly affected by the experimental treatments (2-way ANOVA), although epiphyte biomass on eelgrass leaves was significantly higher under high nutrient loading (0.164 ± 0.013 mg AFDW cm^{-2}) and adult oyster filtration (0.179 ± 0.011 mg AFDW cm^{-2}) compared to low nutrient loading (0.140 ± 0.012 mg AFDW cm^{-2}) and no adult oyster filtration (0.126 ± 0.006 mg AFDW cm^{-2}; $p<0.05$ by nutrient treatment, $p<0.001$ by oyster treatment, 2-way ANOVA). Microzooplankton grazing rates were not significantly different between treatments, and ranged from 2.31 to 2.39 d^{-1} (Table 3). POC/PON data were not available for this experiment.

Experiment 3

Experiment 3 ran from August 22nd to September 6th, 2007. The average temperature in the experimental tanks was 24.56 ± 0.15 °C, the average salinity was 29.73 ± 0.09, and the average dissolved oxygen was 6.16 ± 0.16 mg L^{-1}. The "low nutrient loading" treatment

received an average of 0.134 mmoles N m^{-2} d^{-1} through a 2% d^{-1} exchange with Shinnecock Bay water. The "high nutrient loading" treatment received ambient exchange plus a daily experimental nutrient addition for a total of 5.49 mmoles N m^{-2} d^{-1}. The densities of adult suspension feeders were 43 or 0 clams m^{-2} (*M. mercenaria*). The estimated turnover time from bivalve filtration for the experimental tanks with hard clams was 1.6 days. All tanks in this experiment were stocked with juvenile scallops, juvenile clams, juvenile oysters, juvenile sheepshead minnows, and eelgrass (Table 1).

In this experiment, the high nutrient loading rate (5.49 mmoles N m^{-2} d^{-1}) and the absence of adult hard clams again produced significant increases in total chlorophyll *a* compared to the low nutrient loading rate (0.134 mmoles N m^{-2} d^{-1}) and the presence of adult hard clams (43 individ. m^{-2}) over the course of a 15-day experiment (Fig. 4A; $p<0.01$, 3-way ANOVA). Trends in total chl *a* were paralleled by the >5 μm size fraction of chl *a* which was higher under high nutrient loading ($p<0.01$, 3-way ANOVA) and in the absence of adult clams ($p<0.001$, 3-way ANOVA, Table 3). The <5 μm size fraction of chl *a* was significantly lower in the presence of adult clam filtration ($p<0.01$, 3-way ANOVA, data not shown), but this smaller size fraction was not significantly altered by the nutrient treatments.

Particulate organic nitrogen (PON) was significantly lower in the presence of adult clams (16.1 ± 1.24 μM) compared to the absence of adult clams (38.4 ± 3.37 μM; Table 3; $p<0.05$, 2-way ANOVA). Particulate organic carbon (POC) was affected by both experimental treatments. The levels of POC were higher in the high nutrient loading treatment (249.76 ± 52.82 μM) compared to the low nutrient loading treatment (215.14 ± 35.57 μM; $p<0.05$, 2-way ANOVA), and POC was lower in the presence of adult clams (116.88 ± 8.62 μM) compared to the absence of adult clams (310.35 ± 18.92 μM; Table 3; $p<0.001$, 2-way ANOVA). The molar ratio of

POC:PON was not significantly affected by any of the treatments in Experiment 3 (Table 3). Silicate accumulated in high nutrient loading mesocosms, while a mid-experiment spike in phosphate and ammonium occurred in the mesocosms with high nutrient loading and adult clams (Table 2). Final levels of nitrate, ammonium, and phosphate did not differ among treatments. Microzooplankton grazing rates were not significantly different between treatments, and ranged from 0.45 to 0.73 d^{-1} (Table 3).

Light penetration to the bottom of the mesocosms was higher in the adult bivalve treatment (Fig. 4B; $p<0.001$, 3-way ANOVA) but was not consistently affected by the nutrient treatments. Eelgrass growth was significantly higher in the presence of adult clams, as measured by leaf area productivity (0.569 ± 0.049 cm^2 $shoot^{-1}$ d^{-1}) or mass productivity (1.87 ± 0.17 mg $shoot^{-1}$ d^{-1}; Fig. 5C) compared to the treatments with no adult clams (0.422 ± 0.036 cm^2 $shoot^{-1}$ d^{-1} leaf area productivity, 1.27 ± 0.23 mg $shoot^{-1}$ d^{-1} mass productivity, $p<0.05$, 2-way ANOVA). Epiphyte growth on the eelgrass blades was also significantly denser in the presence of adult clams (0.186 ± 0.017 mg AFDW cm^{-2}) compared to the absence of adult clams (0.146 ± 0.009 mg AFDW cm^{-2}; $p<0.01$, 2-way ANOVA).

Juvenile clams were not significantly affected by any of the treatment factors in the third experiment. However, juvenile oyster and fish growth were both highest in the absence of adult hard clams. Juvenile oysters grew significantly faster in the absence of adult clams (0.257 ± 0.064 mg AFDW d^{-1}) compared to when adult clams were present (0.034 ± 0.067 mg AFDW d^{-1}; Fig. 5A $p<0.05$, 2-way ANOVA). Juvenile sheepshead minnows also grew significantly faster in the absence of adult clams (0.228 ± 0.017 mm d^{-1} $p<0.05$, 2-way ANOVA) compared to treatments with adult clams (0.177 ± 0.022 mm d^{-1}; Fig 5B). The fish growth rates showed an interesting interaction: the presence/absence of adult clams made more of a difference to the

juvenile sheepshead minnows within the high nutrient loading treatment than within the low nutrient treatment (Fig. 5B; $p<0.05$ interaction, 2-way ANOVA). This interaction was present but non-significant for juvenile oysters (Fig. 5A). Differences in juvenile scallop growth rates were not significant (2-way ANOVA), but juvenile scallop mortality was significantly higher in the presence of adult hard clams than in the absence of adult clams (96% with adult clams, 71% without adult clams; $p<0.001$; G-test of independence, data not shown). Juvenile fish and shellfish were not significantly affected by the nutrient loading treatments in this experiment (2-way ANOVA).

Discussion:

Over the course of three mesocosm experiments both enhanced nutrient loading and filtration by adult hard clams significantly affected the growth of juvenile shellfish, juvenile fish, and eelgrass, as well as phytoplankton and light levels in mesocosms. Adult bivalve filtration and nutrient loading were expected to affect eelgrass growth through changes in the density of phytoplankton, which in turn affects the benthic light regime (Newell and Koch 2004, Wall *et al.* 2008). The results of Experiment 3 were consistent with this hypothesis, where a high density of adult hard clams decreased chlorophyll *a* levels and increased light penetration (Fig 4A-B) leading to an increase in eelgrass productivity (Fig 5C). In Experiment 1, adult hard clams decreased chlorophyll *a* (Fig 1) but did not have a significant effect on light or eelgrass, and there were no consistent effects on chl *a*, light, or eelgrass growth for Experiment 2. Eelgrass growth was not significantly affected by nutrient loading in any of the experiments. The density of epiphytes on eelgrass blades was increased by adult bivalve filtration in Expts 2 and 3, and by nutrient loading in Expt 2. Although thick epiphyte growth has been found to have a negative

impact on seagrass in some cases (Duarte 1995), the densities of epiphytes measured in our experiments (0.13-0.19 mg AFDW cm^{-2}) were likely too low to block light at the blade surface (Brush and Nixon 2002).

Growth rates of juvenile fish and shellfish may be decreased by filtration pressure from adult bivalves, which clear food particles from the water column (Rheault and Rice 1996, Zhou *et al.* 2006), or may be increased by high nutrient loading, which may increase the quantity and quality of suspended food particles (Carmichael *et al.* 2004, Carmichael and Valiela 2005). All three experiments had some results consistent with this hypothesis: juvenile clam growth was increased by high nutrient loading in Experiments 1 and 2 (Fig 2A, 3A), juvenile oyster growth was also increased by high nutrient loading in Experiment 1 (Fig 2B), while juvenile oyster growth was decreased by adult bivalve filtration in Experiments 2 and 3 (Fig 3B, 5A), and juvenile fish growth was also decreased by adult bivalve filtration in Experiment 3 (Fig 5B). Although there were no significant growth responses for scallops, juvenile scallop mortality was increased by adult bivalve filtration in Experiment 3.

The results of these experiments demonstrate the strong reliance of juvenile shellfish and finfish growth rates and survival on the short-term dynamics (days to weeks) of food availability as reflected by concentrations of chlorophyll *a*, POC, and PON. In Experiment 1, where nutrient loading had a strong effect on juvenile growth, the molar ratio of POC:PON was significantly reduced by the high nutrient loading treatment (Table 3), suggesting an enrichment of nitrogen in food particles could have contributed to enhanced shellfish growth (Fig 2A-B). Carmichael *et al.* (2004) and Carmichael and Valiela (2005) have interpreted nitrogen-enriched seston as an increase in the quality of food particles available to juvenile bivalves. Although the molar ratio of POC:PON did not change in Experiment 3, the quantities of POC and PON were both

decreased by adult clam filtration (Table 3), with corresponding decreases in the growth rates of juvenile oysters and sheepshead minnows (Fig 5A-B). Although in most cases increased growth rates of shellfish occurred in parallel with increases in whole or size-fractionated chlorophyll *a*, there were also significant treatment-driven changes in chl *a* that did not produce growth responses in juvenile shellfish (Experiments 1 and 3, adult bivalve and nutrient loading treatments, respectively), and in one case, a shellfish growth response that was not mirrored by any changes in chl *a* (juvenile clams, Experiment 2). It is possible that the availability of food particles to the juvenile shellfish was changed by the treatment factors in this experiment despite static levels of chlorophyll *a* between treatments. Microzooplankton grazing rates, as measured by dilution experiments (Landry et *al.* 1995), ranged from 2.3-2.4 d^{-1} in Experiment 2; these were faster than the estimated turnover time from adult oyster filtration of 1.5 d. Such rapid rates of phytoplankton community turnover could mask true food availability to juvenile bivalves and would account for enhanced bivalve growth responses in Experiment 2 in the absence of changes in chl *a*. In Experiment 3, microzooplankton grazing rates were slower (0.4-0.7 d^{-1}), and comparable to the adult clam filtration time of 1.6 d. In contrast to Experiment 2, this experiment had a clear set of treatment-driven chl *a* differences (Fig 4A) and growth differences in response to adult clam filtration (Fig 5A-C).

During these experiments, the treatment factor driving the effects changed from nutrient loading in the first experiment, to combined factors in the second experiment, and finally to exclusively adult bivalve filtration in the third experiment. These differences may partly reflect differences in treatment administered: Experiment 1 had a larger difference in nutrient loading rate between the high nutrient treatment and the control than other experiments, while Experiment 3 had a larger clam density difference between adult clam treatments than

Experiment 1. These results may have also been influenced by seasonal trends: Lower temperatures during the first experiment (17-23° C) may have yielded lower nutrient regeneration rates (Nagata and Kirchman 1992, Miller *et al.* 1995) and low bivalve filtration rate (Kraeuter and Castagna 2001), making external nutrient loading a more important process. Conversely, higher temperatures (23-25° C) for the second and third experiments likely promoted faster bivalve filtration (Kraeuter and Castagna 2001, Weiss *et al.* 2007) and pelagic nutrient regeneration (Nagata and Kirchman 1992, Miller *et al.* 1995). As such, it seems that either nutrient loading or bivalve filtration can structure estuarine food webs, and the relative importance of these factors can switch seasonally or with changing rates of nutrient loading or bivalve filtration.

The densities of adult hard clams used in our Experiments 1 and 3 (8-12 individ. $tank^{-1}$, or 29-43 individ. m^{-2}) are comparable to historic densities of hard clams in Great South Bay (50-100 individ. m^{-2}, Cerrato *et al.* 2004) but are much lower than current densities in NY estuaries (0.4 - 5 individ. m^{-2}, Weiss *et al.* 2007). Similarly, the density of oysters used in Experiment 2 (6 individ. $tank^{-1}$, or 21 individ. m^{-2}) are comparable to historic densities of Eastern oysters in Chesapeake Bay (43-150 individ. m^{-2}), but are much lower than current densities (0.43 individ. m^{-2}; Newell and Koch 2004). The estimated water column turnover times from these densities of adult bivalves were 1.5 - 2.4 days, within the range reported to control algal bloom formation (Cerrato *et al.* 2004, Wall *et al.* 2008). Consistent with this idea, the presence of adult bivalves yielded lower phytoplankton biomass in all three experiments, although the reduction in chl *a* was not significant for Expt 2. Such ecosystem-wide filtration pressure may have been typical of twentieth-century, unexploited natural bivalve populations in Chesapeake Bay (Newell 1988, Newell and Koch 2004) or Great South Bay (McHugh 1991, Cerrato *et al.* 2004). Similarly,

modern high-density bivalve aquaculture may also achieve these ecosystem filtration rates (Nunes *et al.* 2003, Zhou *et al.* 2006). Estuarine management programs may consider bivalve restoration as a management tool to control pelagic algal blooms (Cerrato *et al.* 2004), combat eutrophication (Cerco and Seitzinger 2007), and facilitate the growth of eelgrass (Fig 5C; Peterson and Heck, 2001, Newell and Koch, 2004, Wall *et al.* 2008), although the potential impacts on juvenile shellfish must also be considered.

While enhanced bivalve filtration was beneficial to eelgrass and to some extent epiphytes on eelgrass, they exerted a significantly negative effect on the growth of juvenile fish and shellfish (Fig 3B, 5A-B) and in one case even led to a significant increase in juvenile scallop mortality (Expt 3). Rheault and Rice (1996) placed juvenile Eastern oysters (*C. virginica*) and bay scallops (*A. irradians*) in a compartmented flume and found decreased growth and condition index in the shellfish that were downstream compared to the upstream dense populations. From an initial ambient chlorophyll *a* concentration of 4 to 8 $\mu g\ L^{-1}$, each batch of shellfish decreased chl *a* by 27-35% compared to the upstream compartment (Rheault and Rice 1996). In Experiment 3 of our study, the high density of adult hard clams produced a comparable drop in mean chl *a* levels (Fig. 4A, -37%) compared to the control, and also led to decreased growth of juvenile oysters (Fig 5A) and survival of juvenile scallops. The concentrations of chl *a* in Experiment 3 were relatively high ($25.09 \pm 2.56\ \mu g\ L^{-1}$ with no adult clams; $15.90 \pm 2.20\ \mu g\ L^{-1}$ with adult clams; Table 3); this drop in chlorophyll *a* produced a significant decrease in juvenile oyster growth but not juvenile clam growth. It is likely that juvenile clam food requirements were saturated at a lower chlorophyll *a* concentration than juvenile oyster food requirements (Tenore and Dunstan 1973). These impacts illustrate an eventual trade-off between the benefit and cost of higher ecosystem filtration rates: Despite the benefits to seagrass, high rates of water

column turnover by adult shellfish could serve as a negative feedback on juvenile fish and shellfish populations (Fig 3B, 5A-B) by decreasing food availability (Fig 4A). Such density-dependent food limitation is a common phenomenon within bivalve aquaculture (Rheault and Rice 1996, Zhou *et al.* 2006). The extent to which this may occur within estuarine ecosystems is not well known, but will certainly depend on the species involved and the particular physics and biology of each ecosystem (Newell 2004, Ferreira *et al.* 2008).

Many estuarine management plans have focused on the need to reduce nutrient loads to mitigate the effects of eutrophication (Nixon 1995, Cloern 2001, de Jonge *et al.* 2002). Partly through changes in land use and better sewage treatment, inorganic nutrient levels and/or chlorophyll *a* concentrations have declined in many formerly eutrophic systems, such as the North Sea (Nunnieri *et al.* 2007, Artioli *et al.* 2008), the Dutch Wadden Sea (Philippart *et al.* 2007), Narragansett Bay, RI, USA (Fulweiler *et al.* 2007), Long Island Sound, USA (CTDEP 1991 – 2007), and the Peconic Estuary, NY, USA (SCDHS, 1976-2005). Despite this easing of eutrophication as measured by declines in inorganic nutrient and chlorophyll *a* concentrations, the recovery of estuarine resources in these systems has not been reported. The high nutrient loading rates in our experimental tanks are comparable to measured nutrient loading rates in eutrophic northeast U.S. estuaries (Taylor *et al.* 1999), from which valuable estuarine resources have been lost (Ryther 1989, McHugh *et al.* 1991, Valiela *et al.* 1992). However, there have been reported positive effects of moderate levels of nutrient loading. Weiss *et al.* (2002) and Carmichael *et al.* (2004) both saw increases in shell growth and soft tissue growth of quahogs (hard clams, *Mercenaria mercenaria*) and softshell clams (*Mya arenaria*) over the nutrient gradient from mesotrophic to eutrophic regions of the Waquoit Bay system, MA, USA. This parallels our increase in growth for juvenile hard clams and Eastern oysters from low to high

nutrient loading in Experiment 1 and juvenile clams in Experiment 2. Although Carmichael *et al.* (2004) also saw hypoxia in the systems with the highest nutrient loads, which decreased juvenile clam survival, our well-mixed mesocosms remained normoxic during experiments. In light of this information, our findings suggest that inorganic nutrient loading might be allowed to increase in some relatively oligotrophic and well-mixed coastal systems with increased secondary production of resource species as a positive benefit (Nixon and Buckley 2002). Of course, such potential benefits would need to be considered in light of potentially negative effects of higher nutrient loads in an ecosystem such as hypoxia (Diaz and Rosenberg 2008), loss of seagrass beds (Valiela *et al.* 1992, Dennison *et al.* 1989), and harmful algal blooms (Anderson *et al.* 2008).

Future ecosystem-based management of estuaries will need to simultaneously administer bivalve restoration, control of nutrient loading, conservation of key fishery species, the burgeoning aquaculture industry, and protection of critical habitats such as seagrass meadows and salt marshes. Bivalve filtration and nutrient loading can have significant and complex impacts on the growth of estuarine resources, including eelgrass, finfish, and shellfish. Based on the results of these experiments and other findings, some general conclusions can be drawn. The first is that eelgrass is light-limited in many eutrophic estuaries (Dennison and Alberte 1985, Duarte 1995) and will benefit from proximity to the enhanced filtration of bivalve beds (Wall *et al.* 2008). Additionally, bivalves can benefit seagrasses through enhanced biodeposition (Peterson and Heck 2001, Carroll *et al.* 2008). As such, re-planting of eelgrass beds should focus on areas that have high light penetration and/or are adjacent to existing dense bivalve populations. The second conclusion is based on our finding that juvenile resource bivalves respond to enhanced inorganic nutrient loading with increased growth, and respond with

decreased growth to high densities of adult bivalves. This is likely mediated by food limitation: inorganic nutrients encourage the growth of larger and more nutritious phytoplankton (Wikfors *et al.* 1992, Raven and Kubler 2002) while dense collections of adult bivalves can limit juvenile growth by clearing too many of these food particles (Rheault and Rice 1996, Zhou *et al.* 2006). Clearly, predators (Gosselin and Qian 1997, Polyakov *et al.* 2007) and hypoxia (Breitburg 2002, Altieri and Witman 2006) also exert significant mortalities on juvenile bivalves in the field. However, in absence of hypoxia and differential predation, restoration, re-seeding, and aquaculture of clams, oysters, and scallops are more likely to succeed in areas that have moderate nutrient loading rates. Managers must carefully consider the spacing between aquaculture operations as well as between aquaculture operations and natural bivalve populations.

Acknowledgements

Many people provided field and laboratory support for this project including A. Burson, B. Rodgers, J. Goleski, M. Marcoval, A. Stubler, C. Blankenship, J. Carroll, C. Thomas, S. Brisbin, T. Duffy, T. Davis, F. Koch, S. Talmage, T. Hattenrath, T. Behling, D. Getz, M. Meade, B. Gagliardi, and M. Harke. We thank Evan Ward for guidance in performing clearance rate measurements. Juvenile shellfish were provided by G. Rivara and M. Patricio at Cornell Cooperative Extension, Southold, NY. This project was supported by The Nature Conservancy. This is contribution #### from the School of Marine and Atmospheric Sciences at Stony Brook University.

References:

Altieri AH, Witman JD (2006) Local extinction of a foundation species in a hypoxic estuary: Integrating individuals to ecosystem. Ecology 87:717-730

Anderson DM, Burkholder JM, Cochlan WP, Glibert PM, Gobler CJ, Heil CA, Kudela RM, Parsons ML, Rensel JEJ, Townsend DW, Trainer VL, Vargo GA (2008) Harmful algal blooms and eutrophication: Examining linkages from selected coastal regions of the United States. Harmful Algae 8:39-53

Artioli Y, Friedrich J, Gilbert AJ, McQuatters-Gollop A, Mee LD, Vermaat JE, Wulff F, Humborg C, Palmeri L, Pollehne F (2008) Nutrient budgets for European seas: A measure of the effectiveness of nutrient reduction policies. Marine Pollution Bulletin 56:1609-1617

Barber BJ, Davis CV (1997) Growth and mortality of cultured bay scallops in the Damariscotta River, Maine (USA). Aquaculture International 5:451-460

Bass AE, Malouf RE, Shumway SE (1990) Growth of Northern Quahogs (*Mercenaria mercenaria*) fed on picoplankton. Journal of Shellfish Research 9:299-307

Beck MW, Heck KL, Able KW, Childers DL, Eggleston DB, Gillanders BM, Halpern B, Hays CG, Hoshino K, Minello TJ, Orth RJ, Sheridan PF, Weinstein MR (2001) The identification, conservation, and management of estuarine and marine nurseries for fish and invertebrates. Bioscience 51:633-641

Beukema JJ, Cadee GC (1991) Growth-Rates of the Bivalve *Macoma-balthica* in the Wadden Sea During a Period of Eutrophication - Relationships with Concentrations of Pelagic Diatoms and Flagellates. Marine Ecology-Progress Series 68:249-256

Breitburg D (2002) Effects of hypoxia, and the balance between hypoxia and enrichment, on coastal fishes and fisheries. Estuaries 25:767-781

Bricelj VM, Kuenstner SH (1989) Effects of the "brown tide" on the feeding physiology and growth of bay scallops and mussels. In: Cosper EM (ed) Novel phytoplankton blooms: Causes and impacts of recurrent brown tides and other unusual blooms. Springer, p 491-509

Brush MJ, Nixon SW (2002) Direct measurements of light attenuation by epiphytes on eelgrass Zostera marina. Marine Ecology-Progress Series 238:73-79

Calbet A, Landry MR (2004) Phytoplankton growth, microzooplankton grazing, and carbon cycling in marine systems. Limnology and Oceanography 49:51-57

Carmichael RH, Shriver AC, Valiela I (2004) Changes in shell and soft tissue growth, tissue composition, and survival of quahogs, *Mercenaria mercenaria*, and softshell clams, *Mya arenaria*, in response to eutrophic-driven changes in food supply and habitat. Journal of Experimental Marine Biology and Ecology 313:75-104

Carmichael RH, Valiela L (2005) Coupling of near-bottom seston and surface sediment composition: Changes with nutrient enrichment and implications for estuarine food supply and biogeochemical processing. Limnology and Oceanography 50:97-105

Carroll J, Gobler CJ, Peterson BJ (2008) Resource-restricted growth of eelgrass in New York estuaries: light limitation, and alleviation of nutrient stress by hard clams. Marine Ecology-Progress Series 369:51-62

Cerco CF, Noel MR (2007) Can oyster restoration reverse cultural eutrophication in Chesapeake Bay? Estuaries and Coasts 30:331-343

Cerrato RM, Caron DA, Lonsdale DJ, Rose JM, Schaffner RA (2004) Effect of the northern quahog *Mercenaria mercenaria* on the development of blooms of the brown tide alga *Aureococcus anophagefferens*. Marine Ecology-Progress Series 281:93-108

Cloern JE (2001) Our evolving conceptual model of the coastal eutrophication problem. Marine Ecology-Progress Series 210:223-253

Connecticut Department of Environmental Protection (CTDEP). 1991 – 2007. Long Island Sound Annual Water Quality Data Report. Hartford, CT, USA.

de Jonge VN, Elliott M, Orive E (2002) Causes, historical development, effects and future challenges of a common environmental problem: eutrophication. Hydrobiologia 475:1-19

Dennison WC, Alberte RS (1985) Role of Daily Light Period in the Depth Distribution of *Zostera-marina* (Eelgrass). Marine Ecology-Progress Series 25:51-61

Dennison WC, Marshall GJ, Wigand C (1989) Effects of 'brown tide' shading on eelgrass (*Zostera marina*) distributions. In: Cosper EM, Bricelj VM, Carpenter EJ (eds) Novel phytoplankton blooms: causes and impacts of recurrent brown tides and other unusual blooms. Springer-Verlag, Berlin, p 675–692

Diaz RJ, Rosenberg R (2008) Spreading dead zones and consequences for marine ecosystems. Science 321:926-929

Doering PH, Oviatt CA (1986) Application of Filtration-Rate Models to Field Populations of Bivalves - an Assessment Using Experimental Mesocosms. Marine Ecology-Progress Series 31:265-275

Dolmer P, Frandsen RP (2002) Evaluation of the Danish mussel fishery: suggestions for an ecosystem management approach. Helgoland Mar Res 56:13-20

Duarte CM (1995) Submerged Aquatic Vegetation in Relation to Different Nutrient Regimes. Ophelia 41:87-112

Ferreira JG, Hawkins AJS, Monteiro P, Moore H, Service M, Pascoe PL, Ramos L, Sequeira A (2008) Integrated assessment of ecosystem-scale carrying capacity in shellfish growing areas. Aquaculture 275:138-151

Frechette M, Aitken AE, Page L (1992) Interdependence of Food and Space Limitation of a Benthic Suspension Feeder - Consequences for Self-Thinning Relationships. Marine Ecology-Progress Series 83:55-62

Fulweiler RW, Nixon SW, Buckley BA, Granger SL (2007) Reversal of the net dinitrogen gas flux in coastal marine sediments. Nature 448:180-182

Gabbott PA, Walker AJM (1971) Changes in Condition Index and Biochemical Content of Adult Oysters (*Ostrea-edulis* L)Maintained under Hatchery Conditions. Journal Du Conseil 34:99-&

Gifford S, Dunstan RH, O'Connor W, Roberts T, Toia R (2004) Pearl aquaculture - profitable environmental remediation? Sci Total Environ 319:27-37

Gobler CJ, Lonsdale DJ, Boyer GL (2005) A review of the causes, effects, and potential management of harmful brown tide blooms caused by *Aureococcus anophagefferens* (Hargraves et Sieburth). Estuaries 28:726-749

Gobler CJ, Sanudo-Wilhelmy SA (2001) Temporal variability of groundwater seepage and brown tide blooms in a Long Island embayment. Marine Ecology-Progress Series 217:299-309

Gosselin LA, Qian PY (1997) Juvenile mortality in benthic marine invertebrates. Marine Ecology-Progress Series 146:265-282

Hallegraeff GM (1993) A Review of Harmful Algal Blooms and Their Apparent Global Increase. Phycologia 32:79-99
Hardy CD (1976) A preliminary description of the Peconic Bay Estuary. Report No. 76-4, Marine Sciences Research Center, Stony Brook, NY
Heck KL, Wetstone GS (1977) Habitat Complexity and Invertebrate Species Richness and Abundance in Tropical Seagrass Meadows. Journal of Biogeography 4:135-142
Huang CH, Lin HJ, Huang TC, Su HM, Hung JJ (2008) Responses of phytoplankton and periphyton to system-scale removal of oyster-culture racks from a eutrophic tropical lagoon. Marine Ecology-Progress Series 358:1-12
Ibarraobando SE, Boudouresque CF (1994) An Improvement of the Zieman Leaf Marking Technique for *Zostera-marina* Growth and Production Assessment. Aquat Bot 47:293-302
Irlandi EA, Peterson CH (1991) Modification of Animal Habitat by Large Plants - Mechanisms by Which Seagrasses Influence Clam Growth. Oecologia 87:307-318
Jackson JBC (2001) What was natural in the coastal oceans? Proceedings of the National Academy of Sciences of the United States of America 98:5411-5418
Jackson JBC, Kirby MX, Berger WH, Bjorndal KA, Botsford LW, Bourque BJ, Bradbury RH, Cooke R, Erlandson J, Estes JA, Hughes TP, Kidwell S, Lange CB, Lenihan HS, Pandolfi JM, Peterson CH, Steneck RS, Tegner MJ, Warner RR (2001) Historical overfishing and the recent collapse of coastal ecosystems. Science 293:629-638
Kemp WM, Boynton WR, Adolf JE, Boesch DF, Boicourt WC, Brush G, Cornwell JC, Fisher TR, Glibert PM, Hagy JD, Harding LW, Houde ED, Kimmel DG, Miller WD, Newell RIE, Roman MR, Smith EM, Stevenson JC (2005) Eutrophication of Chesapeake Bay: historical trends and ecological interactions. Marine Ecology-Progress Series 303:1-29
Kraeuter JN, Castagna M (2001) Biology of the Hard Clam, Vol. Elsevier Science, New York
Landry MR, Kirshtein J, Constantinou J (1995) A Refined Dilution Technique for Measuring the Community Grazing Impact of Microzooplankton, with Experimental Tests in the Central Equatorial Pacific. Marine Ecology-Progress Series 120:53-63
Lotze HK, Lenihan HS, Bourque BJ, Bradbury RH, Cooke RG, Kay MC, Kidwell SM, Kirby MX, Peterson CH, Jackson JBC (2006) Depletion, degradation, and recovery potential of estuaries and coastal seas. Science 312:1806-1809
Malone TC (1980) Algal size. In: Morris I (ed) The Physiological Ecology of Phytoplankton. Blackwell, Oxford, p 433-463
McHugh JL (1991) The hard clam fishery past and present. In: Schubel JR, Bell TM, Carter HH (eds) The Great South Bay. SUNY Press
Miller CA, Penry DL, Glibert PM (1995) The impact of trophic interactions on rates of nitrogen regeneration and grazing in Chesapeake Bay. Limnology and Oceanography 40:1005–1011
Nagata T, Kirchman DL (1992) Release of macromolecular organic complexes by heterotrophic marine flagellates. Marine Ecology-Progress Series 83:233–240
Naylor RL, Goldburg RJ, Primavera JH, Kautsky N, Beveridge MCM, Clay J, Folke C, Lubchenco J, Mooney H, Troell M (2000) Effect of aquaculture on world fish supplies. Nature 405:1017-1024
Newell RIE (1988) Ecological changes in Chesapeake Bay, are they the result of overharvesting the eastern oyster (*Crassostrea virginica*)? In: Lynch MP, Krome EC (eds)

Understanding the estuary, Publ 129, Chesapeake Research Consortium, Gloucester Point, VA (also available at www.vims.edu/GreyLit/crc129.pdf)
Newell RIE (2004) Ecosystem influences of natural and cultivated populations of suspension-feeding bivalve molluscs: A review. Journal of Shellfish Research 23:51-61
Newell RIE, Cornwell JC, Owens MS (2002) Influence of simulated bivalve biodeposition and microphytobenthos on sediment nitrogen dynamics: A laboratory study. Limnology and Oceanography 47:1367-1379
Newell RIE, Koch EW (2004) Modeling seagrass density and distribution in response to changes in turbidity stemming from bivalve filtration and seagrass sediment stabilization. Estuaries 27:793-806
Nixon SW (1995) Coastal Marine Eutrophication - a Definition, Social Causes, and Future Concerns. Ophelia 41:199-219
Nixon SW, Buckley BA (2002) "A strikingly rich zone" - Nutrient enrichment and secondary production in coastal marine ecosystems. Estuaries 25:782-796
Nunes JP, Ferreira JG, Gazeau F, Lencart-Silva J, Zhang XL, Zhu MY, Fang JG (2003) A model for sustainable management of shellfish polyculture in coastal bays. Aquaculture 219:257-277
Nunneri C, Windhorst W, Turner RK, Lenhart H (2007) Nutrient emission reduction scenarios in the North Sea: An abatement cost and ecosystem integrity analysis. Ecological Indicators 7:776-792
Officer CB, Smayda TJ, Mann R (1982) Benthic Filter Feeding - a Natural Eutrophication Control. Marine Ecology-Progress Series 9:203-210
Orth RJ, Carruthers TJB, Dennison WC, Duarte CM, Fourqurean JW, Heck KL, Hughes AR, Kendrick GA, Kenworthy WJ, Olyarnik S, Short FT, Waycott M, Williams SL (2006) A global crisis for seagrass ecosystems. Bioscience 56:987-996
PEPCCMP (2003) Peconic Estuary Program Comprehensive Conservation and Management Plan.
Parsons TR, Y. Maita, C.M. Lalli (1984) A manual of chemical and biological methods for seawater analysis., Vol. Pergamon Press, Oxford
Peterson BJ, Heck KL (2001) An experimental test of the mechanism by which suspension feeding bivalves elevate seagrass productivity. Marine Ecology-Progress Series 218:115-125
Philippart CJM, Beukema JJ, Cadee GC, Dekker R, Goedhart PW, van Iperen JM, Leopold MF, Herman PMJ (2007) Impacts of nutrient reduction on coastal communities. Ecosystems 10:95-118
Polyakov O, Kraeuter JN, Hofmann EE, Buckner SC, Bricelj VM, Powell EN, Klinck JM (2007) Benthic predators and northern quahog (=hard clam) (*Mercenaria mercenaria* Linnaeus, 1758) populations. Journal of Shellfish Research 26:995-1010
Raven JA, Kubler JE (2002) New light on the scaling of metabolic rate with the size of algae. Journal of Phycology 38:11-16
Rheault RB, Rice MA (1996) Food-limited growth and condition index in the eastern oyster, *Crassostrea virginica* (Gmelin 1791), and the bay scallop, *Argopecten irradians irradians* (Lamarck 1819). Journal of Shellfish Research 15:271-283
Riisgard HU (2001) On measurement of filtration rates in bivalves - the stony road to reliable data: review and interpretation. Marine Ecology-Progress Series 211:275-291

Ruesink JL, Lenihan HS, Trimble AC, Heiman KW, Micheli F, Byers JE, Kay MC (2005) Introduction of non-native oysters: Ecosystem effects and restoration implications. Annual Review of Ecology, Evolution, and Systematics 36:643-689

Ryther JH (1989) Historical perspective of phytoplankton blooms on Long Island and the green tides of the 1950's.. *In* E. M. Cosper, V. M. Bricelj, and E. J. Carpenter, (eds.), Novel Phytoplankton Blooms. Coastal and Estuarine Studies. Springer Verlag, New York, p 375–381

SCDHS (1976-2005) Annual report on water quality in Suffolk County's coastal waters. Suffolk County (NY) Department of Health Services, Office of Ecology

Samson JC, Shumway SE, Weis JS (2008) Effects of the toxic dinoflagellate, *Alexandrium fundyense* on three species of larval fish: a food-chain approach. Journal of Fish Biology 72:168-188

Sharp JH (1974) Improved Analysis for Particulate Organic Carbon and Nitrogen from Seawater. Limnology and Oceanography 19:984-989

Shriver AC, Carmichael RH, Valiela I (2002) Growth, condition, reproductive potential, and mortality of bay scallops, *Argopecten irradians*, in response to eutrophic-driven changes in food resources. Journal of Experimental Marine Biology and Ecology 279:21-40

Smaal AC, Prins TC (1993) The uptake of organic matter and the release of inorganic nutrients by bivalve suspension feeder beds. In: Dame RF (ed) Bivalve filter feeders in estuarine and coastal ecosystem processes. Springer-Verlag, Berlin, p 271–298

Sokal RR, Rohlf FJ (1995) Biometry. WH Freeman, New York

Sunda WG, Graneli E, Gobler CJ (2006) Positive feedback and the development and persistence of ecosystem disruptive algal blooms. Journal of Phycology 42:963-974

Taylor DI, Nixon SW, Granger SL, Buckley BA (1999) Responses of coastal lagoon plant communities to levels of nutrient enrichment: A mesocosm study. Estuaries 22:1041-1056

Tenore KR, Dunstan WM (1973) Comparison of Feeding and Biodeposition of 3 Bivalves at Different Food Levels. Mar Biol 21:190-195

Valderrama JC (1981) The Simultaneous Analysis of Total Nitrogen and Total Phosphorus in Natural-Waters. Marine Chemistry 10:109-122

Valiela I, Foreman K, Lamontagne M, Hersh D, Costa J, Peckol P, Demeoandreson B, Davanzo C, Babione M, Sham CH, Brawley J, Lajtha K (1992) Couplings of Watersheds and Coastal Waters - Sources and Consequences of Nutrient Enrichment in Waquoit Bay, Massachusetts. Estuaries 15:443-457

Vanstralen MR, Dijkema RD (1994) Mussel Culture in a Changing Environment - the Effects of a Coastal Engineering Project on Mussel Culture (*Mytilus-edulis*-L) in the Oosterschelde Estuary (Sw Netherlands). Hydrobiologia 283:359-379

Wall CC, Peterson BJ, Gobler CJ (2008) Facilitation of seagrass *Zostera marina* productivity by suspension-feeding bivalves. Marine Ecology-Progress Series 357:165-174

Weiss ET, Carmichael RH, Valiela I (2002) The effect of nitrogen loading on the growth rates of quahogs (*Mercenaria mercenaria*) and soft-shell clams (*Mya arenaria*) through changes in food supply. Aquaculture 211:275-289

Weiss MB, Curran PB, Peterson BJ, Gobler CJ (2007) The influence of plankton composition and water quality on hard clam (*Mercenaria mercenaria* L.) populations across Long Island's south shore lagoon estuaries (New York, USA). Journal of Experimental Marine Biology and Ecology 345:12-25

Wilson RE, Wong KC, Carter HH (1991) Aspects of circulation and exchange in Great South Bay. In: Schubel JR, Bell TM Carter HH (eds) The Great South Bay. SUNY Press, Albany, NY, p 33–42

Wikfors GH, Ferris GE, Smith BC (1992) The Relationship between Gross Biochemical-Composition of Cultured Algal Foods and Growth of the Hard Clam, *Mercenaria-Mercenaria* (L). Aquaculture 108:135-154

Zhou Y, Yang HS, Zhang T, Liu SL, Zhang SM, Liu Q, Xiang JH, Zhang FS (2006) Influence of filtering and biodeposition by the cultured scallop *Chlamys farreri* on benthic-pelagic coupling in a eutrophic bay in China. Marine Ecology-Progress Series 317:127-141

			Experiment 1	Experiment 2	Experiment 3
Stocking densities of response organisms (n = # per tank)	**Juvenile bivalves**	***M. mercenaria***	10	20	10
		C. virginica	15	10	10
		A. irradians	0	0	10
	Juvenile fish	***C. variegatus***	0	0	10
	Eelgrass shoots	***Z. marina***	16	16	16
Experimental conditions	**Adult bivalve species**		*M.mercenaria*	*C. virginica*	*M. mercenaria*
	Density of adult bivalves	**+ B**	29 m^{-2}	21 m^{-2}	43 m^{-2}
		- B	0	0	0
	Estimated filtration time from + B treatment		2.4 d	1.5 d	1.6 d
	Exchange with ambient water		1% d^{-1}	2% d^{-1}	2% d^{-1}
	Nutrient loading rate (mmoles N m^{-2} d^{-1})	**+ N**	10.70	5.75	5.49
		- N	0.065	0.255	0.134

Table 1. Stocking densities of response organisms and summary of experimental conditions. Treatment abbreviations are "+N" or "-N" for high or low nutrient loading, and "+B" or "-B" for presence or absence of adult bivalves. A total of 16 tanks were used for each 2 x 2 factorial experiment with n = 4 tanks per treatment combination.

		Ammonium								Phosphate							
	day	-N/+B		-N/-B		+N/+B		+N/-B		-N/+B		-N/-B		+N/+B		+N/-B	
Expt 1	0	0.62 ±	0.45	0.62 ±	0.45	0.62 ±	0.45	0.62 ±	0.45	0.30 ±	0.07	0.30 ±	0.07	0.30 ±	0.07	0.30 ±	0.07
	7	9.16 ±	3.28	5.17 ±	0.66	58.85 ±	7.78	45.32 ±	6.97	0.61 ±	0.14	0.28 ±	0.08	2.88 ±	0.33	2.30 ±	0.12
	13	16.01 ±	6.92	5.43 ±	2.37	80.94 ±	11.71	52.43 ±	5.92	0.96 ±	0.39	0.32 ±	0.05	2.96 ±	0.49	1.95 ±	0.42
Expt 2	1	4.6 ±	2.02	0.32 ±	0.15	5.84 ±	1.66	2.22 ±	0.79	1.47 ±	0.38	0.58 ±	0.07	1.36 ±	0.18	1.24 ±	0.27
	8	2.34 ±	1.02	1.13 ±	0.74	10.87 ±	3.96	2.60 ±	1.32	4.36 ±	0.60	3.34 ±	0.66	4.64 ±	0.41	4.11 ±	0.53
	15	3.82 ±	1.46	2.10 ±	1.82	4.63 ±	1.55	4.34 ±	1.46	4.33 ±	0.06	4.26 ±	0.23	4.34 ±	0.07	4.33 ±	0.06
Expt 3	0	4.33 ±	0.19	4.33 ±	0.19	4.33 ±	0.19	4.33 ±	0.19	1.87 ±	0.06	1.87 ±	0.06	1.87 ±	0.06	1.87 ±	0.06
	7	2.52 ±	1.16	1.00 ±	0.11	15.46 ±	5.75	5.85 ±	0.94	1.72 ±	0.41	0.71 ±	0.09	3.58 ±	0.7	1.14 ±	0.99
	15	6.67 ±	0.44	6.37 ±	0.20	6.57 ±	0.39	7.17 ±	0.25	4.71 ±	0.55	5.26 ±	0.66	4.32 ±	0.15	4.17 ±	0.05

		Nitrate								Silicate							
	day	-N/+B		-N/-B		+N/+B		+N/-B		-N/+B		-N/-B		+N/+B		+N/-B	
Expt 1	0	5.03 ±	0.39	5.03 ±	0.39	5.03 ±	0.39	5.03 ±	0.39	26.84 ±	3.25	26.84 ±	3.25	26.84 ±	3.25	26.84 ±	3.25
	7	8.35 ±	1.13	5.89 ±	0.16	7.02 ±	0.09	7.99 ±	0.26	38.39 ±	1.17	34.45 ±	3.30	33.99 ±	2.85	35.08 ±	2.87
	13	2.79 ±	0.37	2.11 ±	0.66	4.50 ±	0.83	5.27 ±	0.80	49.92 ±	5.47	50.00 ±	6.90	50.73 ±	3.48	39.56 ±	9.14
Expt 2	1	0.71 ±	0.49	0.49 ±	0.13	0.51 ±	0.20	1.23 ±	0.66	20.92 ±	2.16	14.92 ±	1.24	19.63 ±	2.92	22.63 ±	2.45
	8	0.68 ±	0.13	1.23 ±	0.44	3.36 ±	1.45	0.74 ±	0.26	30.12 ±	5.17	42.29 ±	6.63	39.39 ±	8.70	30.80 ±	3.06
	15	3.08 ±	0.07	3.76 ±	0.48	3.17 ±	0.22	3.08 ±	0.20	27.63 ±	0.38	27.09 ±	0.11	28.48 ±	0.66	27.97 ±	0.73
Expt 3	0	4.19 ±	0.55	4.19 ±	0.55	4.19 ±	0.55	4.19 ±	0.55	32.68 ±	0.71	32.68 ±	0.71	32.68 ±	0.71	32.68 ±	0.71
	7	1.83 ±	0.72	1.22 ±	0.24	1.32 ±	0.70	1.21 ±	0.62	33.88 ±	1.75	32.43 ±	2.05	33.95 ±	2.37	32.17 ±	1.30
	15	2.52 ±	0.14	4.07 ±	0.37	2.99 ±	0.41	3.28 ±	0.57	47.01 ±	9.69	45.9 ±	5.10	37.27 ±	2.87	32.72 ±	0.50

Table 2. Levels of inorganic nutrients. Inorganic nutrients ammonium (NH_4^+), phosphate (PO_4^{-3}), nitrate (NO_3^-), and silicate ($Si(OH_4)$). Values in µM are mean ± SE of n = 4 experimental tanks for each treatment and timepoint. Treatment abbreviations are "+N" or "-N" for high or low nutrient loading, and "+B" or "-B" for presence or absence of adult bivalves. Identical values for day 0 of experiments 1 and 3 reflect single nutrient readings taken from source water for each experiment.

		whole chl *a* µg L^{-1}	>5 µm chl *a* µg L^{-1}	POC µM	PON µM	POC:PON	Microzooplankton grazing rate d^{-1}
Expt 1	**-N/+B**	6.60 ± 0.91	4.92 ± 0.75	244.00 ± 44.06	27.21 ± 3.83	8.85 ± 0.33	no data
	-N/-B	8.39 ± 1.18	5.58 ± 0.80	150.46 ± 13.55	13.34 ± 2.09	11.40 ± 0.77	available
	+N/+B	7.72 ± 0.95	5.44 ± 0.83	200.40 ± 26.71	23.13 ± 3.66	8.81 ± 0.39	
	+N/-B	9.38 ± 1.19	6.21 ± 0.84	184.90 ± 15.63	19.25 ± 1.62	9.62 ± 0.23	
Expt 2	**-N/+B**	4.45 ± 0.72	2.63 ± 0.44	no data	no data	no data	2.36 ± 0.52
	-N/-B	4.59 ± 0.64	4.33 ± 1.00	available	available	available	2.39 ± 0.63
	+N/+B	5.71 ± 1.25	2.62 ± 0.43				2.36 ± 0.45
	+N/-B	7.52 ± 1.11	4.47 ± 0.88				2.31 ± 0.53
Expt 3	**-N/+B**	14.15 ± 2.61	8.96 ± 2.30	120.53 ± 18.73	16.05 ± 2.55	7.65 ± 0.89	0.55 ± 0.32
	-N/-B	21.76 ± 3.25	22.58 ± 4.23	272.08 ± 15.24	32.77 ± 3.72	8.42 ± 0.54	0.45 ± 0.07
	+N/+B	19.92 ± 3.64	12.40 ± 3.69	113.24 ± 2.66	16.31 ± 1.07	7.00 ± 0.46	0.73 ± 0.19
	+N/-B	29.00 ± 3.77	31.95 ± 5.76	348.62 ± 9.64	43.95 ± 3.42	8.00 ± 0.47	0.63 ± 0.17

Table 3. Levels of chlorophyll *a*, POC, PON, and microzooplankton grazing rates. Values are mean ± SE of n = 4 experimental tanks for each treatment combination in each experiment. Treatment abbreviations are "+N" or "-N" for high or low nutrient loading, and "+B" or "-B" for presence or absence of adult bivalves. Values of >5 µm chl *a* that are greater than whole chl *a* for Expt 3 reflect plankton communities where virtually all chl *a* is in the >5 µm size-fraction.

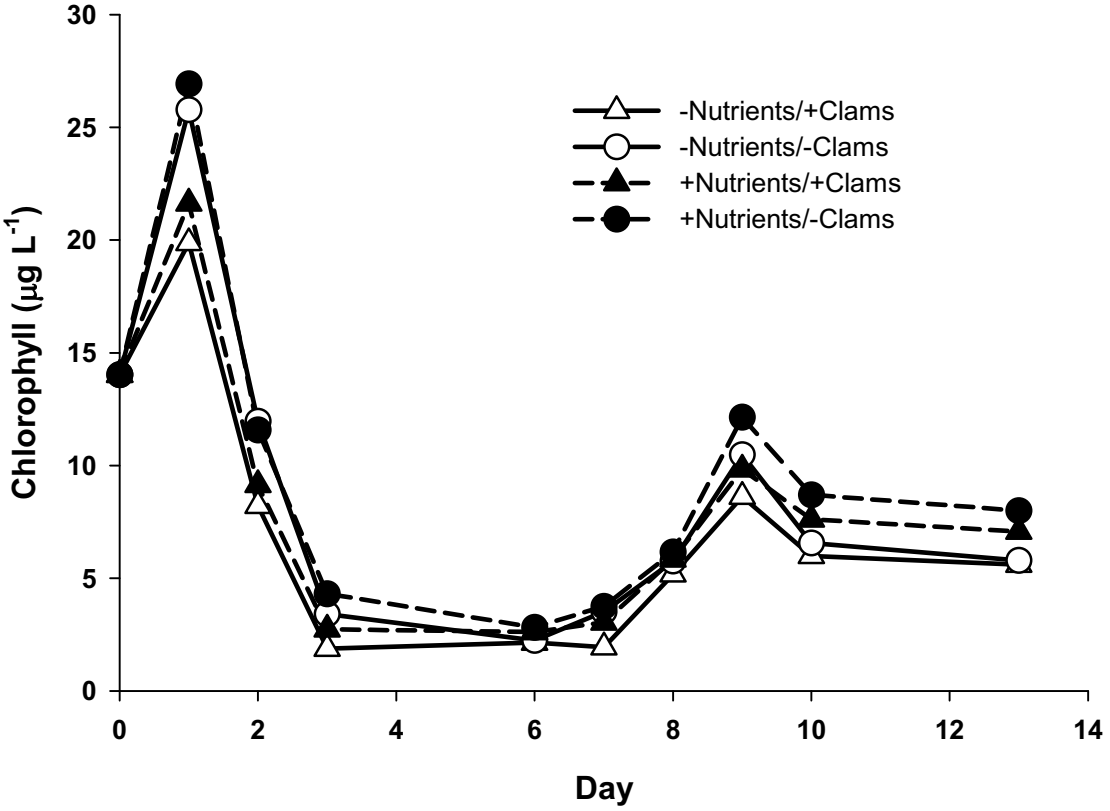


Figure 1: Chlorophyll *a* dynamics in Experiment 1. Data points represent the mean (n=4) for each of the treatment combinations. Error bars are not presented for the sake of visual clarity. The mean relative standard deviation of measurements was 19.8% during the experiment.

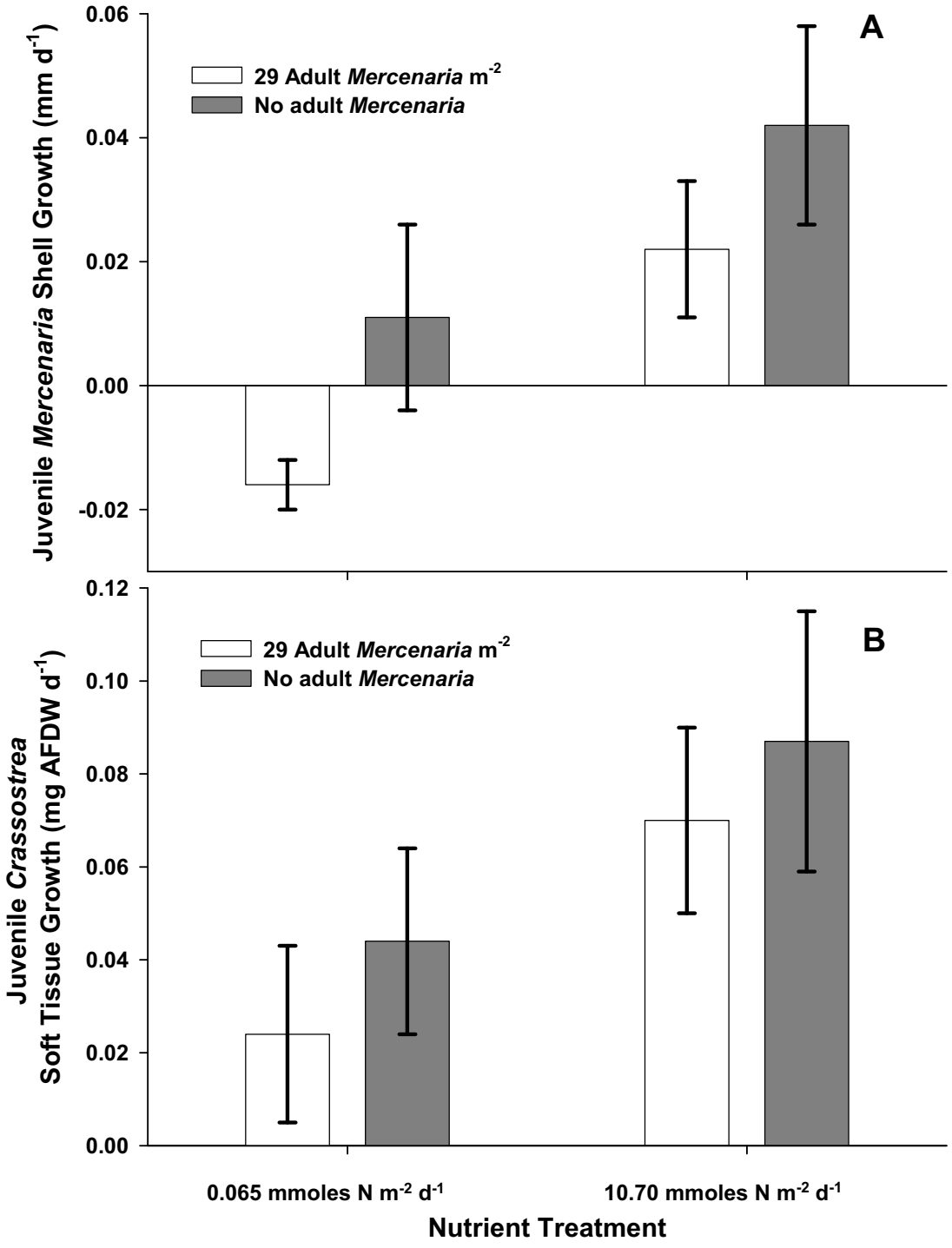


Figure 2: Growth responses from Experiment 1 for (A) juvenile *Mercenaria mercenaria* and (B) juvenile *Crassostrea virginica*. Bars are means ± SE. Slightly negative shell growth for juvenile *M. mercenaria* is within measurement errors of zero.

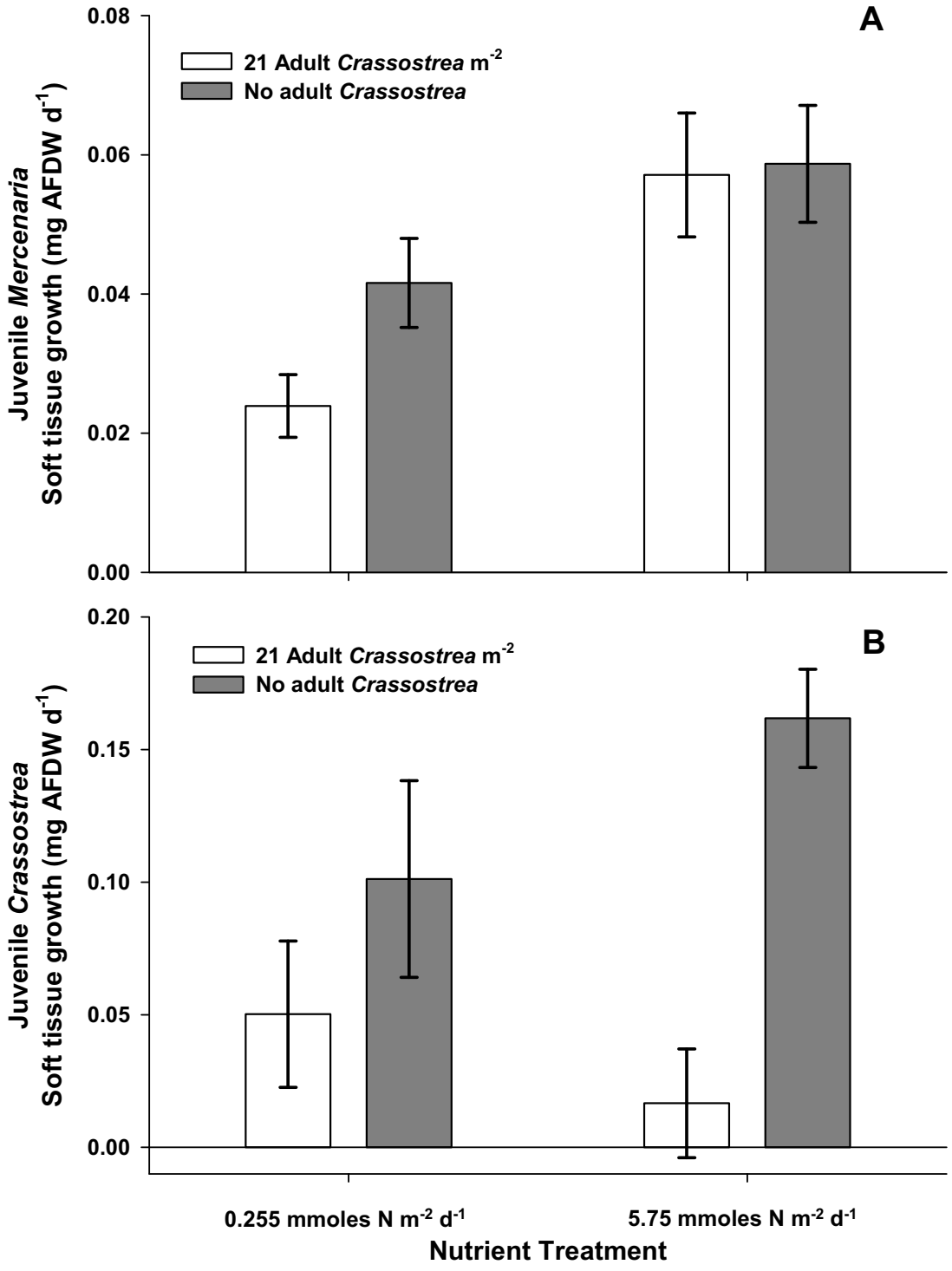


Figure 3: Growth responses from Experiment 2 for (A) juvenile *Mercenaria mercenaria* and (B) juvenile *Crassostrea virginica*. Bars are means ± SE.

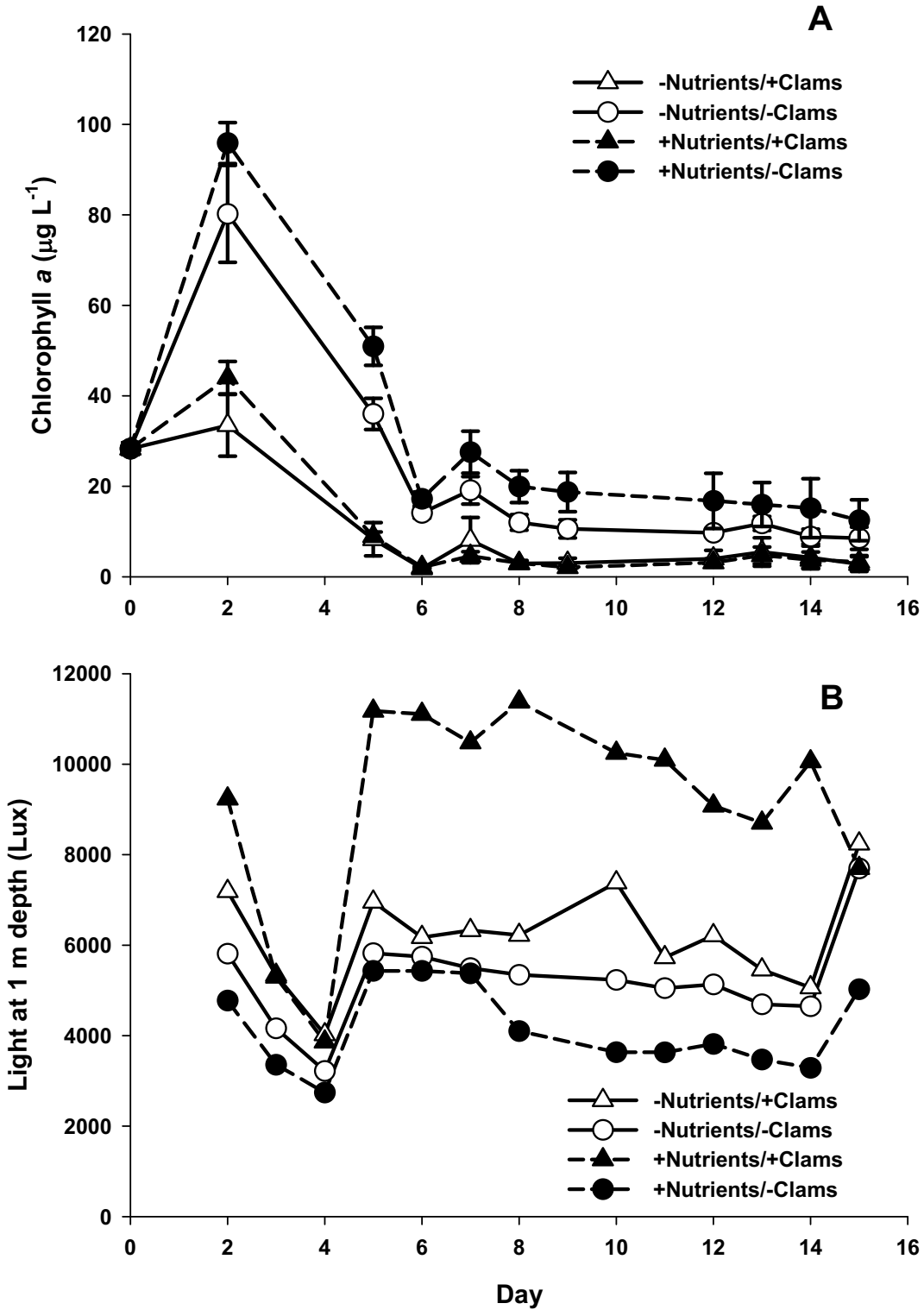


Figure 4: Chlorophyll *a* (A) and light dynamics (B) in Experiment 3. Data points represent the mean (n=4) for each of the treatment combinations. Error bars for light levels are not presented for the sake of visual clarity. The mean relative standard deviation of light measurements was 37.9% during the experiment.

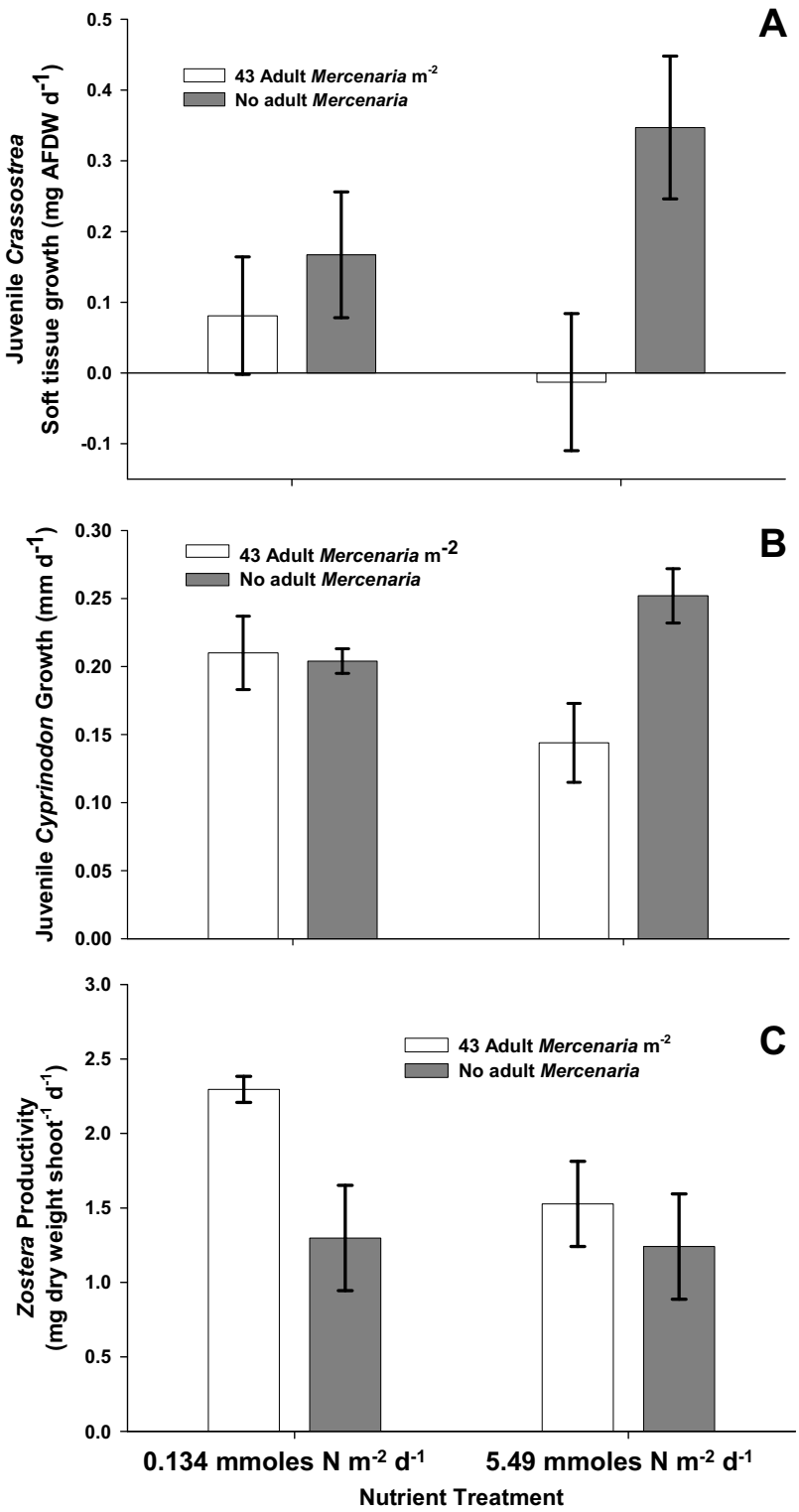


Figure 5: Growth responses from Experiment 3 for (A) juvenile *Crassostrea virginica,* (B) juvenile *Cyprinodon variegatus,* and (C) *Zostera marina.* Bars are means ± SE.

Chapter III:

The facilitation of seagrass (*Zostera marina*) productivity by suspension-feeding bivalves in an experimental setting

This work was begun prior to support from The Nature Conservancy. However, support from the Nature Conservancy allowed the final data analyses and writing of this study to be completed. Additionally, the findings of this study informed the directions taken in chapters I and II of this report.

Key words: *Zostera marina, Crassostrea virginica, Mercenaria mercenaria, Mytilus edulis,* seagrass, eelgrass, eutrophication, bivalves, suspension feeders, mesocosms

ABSTRACT: Seagrasses and suspension feeders are both critical ecosystem engineers in estuaries. Seagrass beds are important structural habitats and suspension feeders, when abundant, can regulate phytoplankton densities. Furthermore, there may be mutual facilitation of growth and recruitment between seagrasses and suspension-feeding bivalves. In a series of mesocosm experiments, the effects of environmentally realistic densities of three different suspension-feeding bivalves (*Mercenaria mercenaria*, *Crassostrea virginica*, *Mytilus edulis*) on the growth of eelgrass (*Zostera marina*) in a eutrophied environment were examined. Experimental treatments with bivalves consistently yielded significantly lower chlorophyll *a* concentrations ($p < 0.05$), and most bivalve treatments also showed significant increases in light penetration ($p < 0.05$). Eelgrass productivity was measured by leaf area growth, and varied from 0.318 ± 0.018 cm^2 $shoot^{-1}$ d^{-1} to 0.832 ± 0.036 cm^2 $shoot^{-1}$ d^{-1} (mean ± SE); leaf area productivity was always significantly higher (on average, 48 ± 9.3% higher) in the treatments with the highest density of bivalves compared to a control without bivalves ($p < 0.05$). The data indicate that clearance of the water column and the subsequent increase in light penetration was the primary mechanism by which suspension-feeding bivalves facilitated the growth of eelgrass. These findings suggest that healthy populations of suspension-feeding bivalves can mitigate the effects of estuarine eutrophication and can facilitate the growth of seagrass in degraded, light-limited habitats.

INTRODUCTION

Estuaries and other coastal ecosystems have suffered multiple anthropogenic insults during the past century including pollution, eutrophication, overfishing of fish and shellfish, introduction of invasive species, and loss of key habitats, such as seagrass beds (Valiela et al., 1992; Nixon, 1995; Newell and Koch, 2004; Jackson et al 2001; Lotze et al., 2006; Valiela, 2006). Seagrass beds are a valuable habitat in many temperate and tropical estuaries, providing structural habitat complexity (Heck and Wetstone, 1977), damping waves and trapping sediment (Newell and Koch, 2004), modifying the sedimentary environment (Reise, 2002), providing a settlement site for juvenile bivalves (Bologna et al., 2005), and furnishing benthic primary production. Due to the array of ecological services provided by seagrass beds, they should be considered "ecosystem engineers" (Reise, 2002; Bruno et al., 2003).

Eutrophication of coastal waters can have a multitude of adverse impacts on affected ecosystems, including nuisance algal blooms, hypoxia, and the subsequent loss of marine life and habitats (Nixon, 1995; de Jonge et al., 2002). Dense phytoplankton blooms or macroalgal growth resulting from eutrophication can reduce light penetration to the benthos and shade seagrass beds (Valiela et al., 1992; Duarte, 1995; Hauxwell et al., 2001; Gobler et al 2005). Anthropogenic nutrient loading can also increase seagrass epiphyte loads which in turn decrease the quantity and quality of light at leaf surfaces and subsequently decrease seagrass productivity (Duarte, 1995). Newell and Koch (2004), through a combination of modeling and field studies in Chesapeake Bay, have shown that the seagrass *Ruppia maritime*, which does not grow deeper than 3 m under optimal conditions, is very sensitive to decreases in light penetration due to algal blooms or

resuspended sediment. Other experimental studies have demonstrated the decline of *Zostera marina* in response to nutrients (Taylor et al., 1999; Bintz et al., 2003), light reduction (Bintz and Nixon, 2001), and epiphyte load (Brush and Nixon, 2002).

One guild of species that might facilitate the growth of seagrass is suspension feeding bivalves. Various species of bivalves live in, on, or near seagrass beds, and the filtration provided by bivalves has the potential to control eutrophication (Officer et al., 1982) and significantly affect carbon cycling (Doering et al., 1986). By depositing solid, nutrient rich fecal material and pseudo-feces, bivalves mediate a flux of organic matter and nutrients out of the water column and into the sediments (Smaal and Prins, 1993). This nutrient enrichment of sediments can increase the growth of seagrass, since seagrasses absorb most of their nutrients from the sediment through their roots and not from the water column (Peterson and Heck, 1999). Reusch et al. (1994) and Peterson and Heck (1999, 2001) showed that *Mytilus edulis* and *Modiolus americanus*, respectively, could increase seagrass productivity through sediment nutrient enrichment. By clearing algal populations and other suspended particles from the water column, bivalves may serve as a control on marine (Cerrato et al., 2004) and freshwater (Heath et al., 1995) algal blooms. While the work of Cerrato et al. (2004) and the modeling efforts of Newell and Koch (2004) suggest that bivalve filtration could increase ambient light and perhaps enhance seagrass productivity, no previous study has directly examined this relationship.

During the past century, many estuarine bivalve populations have suffered from overharvesting and habitat loss (Jackson, 2001; Lotze et al., 2006), an occurrence which could have secondary negative impacts on seagrass beds. Since bivalves may serve as a natural control on eutrophication (Officer et al., 1982), the loss of these populations could

result in decreases in light reaching the benthos, a factor which often limits eelgrass productivity in estuaries (Dennison and Alberte, 1985; Bintz and Nixon, 2001). Great South Bay, a shallow estuary on Long Island's south shore, has experienced an increase in eutrophication and frequent algal blooms, including harmful "brown tides" (*Aureococcus anophagefferens*; Gobler et al., 2005). These changes were concurrent with the loss of seagrass beds (*Zostera marina*; Dennison et al., 1989) and the removal of the dominant suspension feeding bivalve (*Mercenaria mercenaria*; Cerrato et al., 2004). Similarly, in Chesapeake Bay the loss of oyster populations, *Crassostrea virginica*, has been hypothesized to have contributed to the demise of *Z. marina* in this system (Jackson, 2001; Newell and Koch, 2004; Kemp et al., 2005; Lotze et al., 2006).

For this study, the effects of various suspension feeding bivalves on the growth of the seagrass, *Zostera marina*, were examined. A eutrophied system was simulated by loading nutrients to mesocosms containing various combinations of seagrass and bivalves. These experiments were designed to test the hypothesis that, in a eutrophied estuary, algal biomass would decrease and light penetration and seagrass productivity would increase as a function of bivalve filtration pressure.

MATERIAL AND METHODS

Five mesocosm experiments were carried out at the Stony Brook - Southampton Marine Science Center on Old Fort Pond in Southampton, New York from 18 May 2006 to 10 October 2006. Old Fort Pond exchanges tidally with Shinnecock Bay, one of the major Long Island south shore estuaries. The experiments were carried out in a series of 300 L polyethylene tanks (Nalgene©; depth 122 cm, inside diameter 60 cm), which have

been used successfully in the past to examine the impacts of filer-feeding bivalves on pelagic algal communities (Cerrato et al., 2004). Prior to each experiment, all tanks were scrubbed, rinsed with fresh water, and then filled with seawater from Old Fort Pond. The mesocosms were ~90% immersed in Old Fort Pond to maintain a uniform ambient temperature. Small aquarium pumps (Rio® 180 Mini, pumping rate: 456 L h^{-1}) were added to mix the water column of each mesocosm, but were suspended only a few centimeters below the surface to minimize re-suspension of sediments or biodeposits. Measurements taken at the start of each experiment and every 1 -2 days during experiments included temperature, salinity, dissolved oxygen, chlorophyll *a*, and light attenuation. Surface and bottom readings of temperature and salinity during experiments confirmed that aquarium pumps kept the mesocoms well-mixed during experiments. Chlorophyll *a* (chl *a*) was measured by filtering mesocosm samples onto replicated GF/F filters and 5 μm polycarbonate filters, freezing and extracting in acetone, and measuring fluorescence with a Turner Trilogy fluorometer (Parsons et al., 1984). Light was measured using a Li Cor LI-193 spherical underwater quantum sensor, and the light attenuation coefficient, K_d, was calculated from incoming irradiance and light at the bottom of the mesocosm using the following formula:

$$K_d = -\ln(\text{irradiance at depth}/\text{incoming irradiance})/z$$

To stimulate anthropogenic nutrient loading, all mesocosms received daily additions of ammonium (10 μM final concentration) and orthophosphate (0.625 μM final concentration), a nutrient loading rate which mimics rates found within more eutrophic regions of Long Island (Gobler and Boneillo, 2003).

Each mesocosm in all of the experiments contained a weighted plastic planter with clean sand and 12 shoots of the seagrass *Zostera marina*. *Zostera* shoots, 20-30 cm long, were harvested from eastern Shinnecock Bay on the day that each experiment commenced. Eelgrass was sorted to remove reproductive shoots, rinsed in seawater, separated into individual shoots with a segment of the attached rhizome, and marked with a small pinhole at the top of the sheath using an 18 gauge needle, according to the method of Zieman (1974). Twelve marked shoots were randomly assigned to each mesocosm, gently buried in the planter, making sure the roots were intact and covered with sand, and the planters were carefully lowered to the bottom of the mesocosm.

Hard clams (*M. mercenaria*) and oysters (*C. virginica*) were locally harvested, and obtained from seafood markets while blue mussels (*M. edulis*) were collected by hand from Shinnecock Bay. Prior to each experiment, all bivalves were placed in a flowing seawater table for approximately 24 hr to acclimate to the temperature and salinity of Old Fort Pond. To eliminate any impact that the biodeposits might have on elevating productivity, bivalves and eelgrass shoots were separated by plastic dividers within the planter trays. Hard clams were partially buried with the siphon facing up, while mussels and oysters were simply placed on top of the planter.

At the end of each experiment, the planters with their seagrass shoots and bivalves were carefully removed from each mesocosm. Seagrass shoots that detached from the planter during the course of the experiment were not collected for further analysis. In the laboratory, the daily aboveground production and leaf epibiont biomass ($shoot^{-1}$) were determined. Seagrass shoots were collected and their growth was determined. Seagrass productivity was calculated for each experiment as leaf area productivity ($cm^2\ shoot^{-1}\ d^{-1}$)

based on the growth of new leaf material from the shoots in each mesocosm (Zieman, 1974). Epibiont mass was determined by scraping fouling organisms and algae form each leaf then drying them to a constant mass (± 0.01 mg) in an oven at 70^{o}C.

The bivalves were retained after each experiment for the determination of lengths, width, heights, and ash free dry weights. Twelve individuals were randomly selected for a clearance rate measurement using one of the methods outlined by Riisgard (2001). The 12 bivalves were placed in 1 L containers filled with water from one of the control mesocosm tanks which typically had high levels of chlorophyll a (> 20 µg L^{-1}). Experiments commenced when individuals were open and filtering. Chl *a* samples taken before filtration and after a known length of time yielded a clearance rate for each individual according to the formula:

Clearance rate = (volume / time) * ln (initial chl *a* / final chl *a*)

The twelve individuals that were used for the clearance rate measurement were then shucked, dried at 70°C, weighed, combusted at 450°C, and weighed again to determine ash-free dry weights of their tissues. These weights were used to normalize the clearance rates to tissue weight rather than to individual.

A "community" clearance rate for each mesocosm was estimated from these data using the average individual clearance rate and the number of individuals in the tank. An estimated turnover time for the entire tank volume to pass through the bivalves was calculated for each tank by dividing the tank volume by this community clearance rate.

Experiment 1

Experiment 1 was carried out for 18 days from 18 May to 5 June using the hard clam *Mercenaria mercenaria*. There were two experimental treatments with 16 *M. mercenaria* per tank for a density of 57 individ. m^{-2} and a control treatment with no bivalves added (n = 4 for each treatment). This density is comparable to historical densities of 53-105 clams m^{-2} for Great South Bay (Cerrato et al., 2004). Modern densities of *M. mercenaria* in Great South Bay are two magnitudes of order lower ranging from 0.5 to 2 individuals m^{-2} (B. Peterson, pers obs). For all experiments, replicates for each treatment were placed among the array of mesocosms using a randomized blocks design (Sokal & Rohlfe, 1995) to minimize any effects due to placement of the mesocosm tank.

Experiment 2

Experiment 2 was carried out for 12 days from 7 June to 19 June, also with hard clams. There were two experimental treatments with 4 and 8 *M. mercenaria* added for densities of 14 and 29 individ. m^{-2}, respectively, and one control treatment (n = 4 for each treatment).

Experiment 3

Experiment 3 was carried out for 8 days from 20 June to 28 June, this time with the oyster *Crassostera virginica*. The control treatment had no oysters while the two experimental treatments had 2 and 4 oysters added for densities of 7 and 14 individ. m^{-2}, respectively (n = 4 for each treatment). These densities are much higher than modern densities of oysters in Chesapeake Bay, estimated by Newell & Koch (2004) to be 0.43 individ. m^{-2}, but lower than historical densities which are estimated to have been as low

as 43 individ. m^{-2} or as high as 150 individ. m^{-2} in a dense oyster reef habitat (Newell & Koch, 2004).

Experiment 4

Experiment 4 was carried out for 8 days from 6 July to 14 July. Only one experimental treatment of 1 oyster per mesocosm (4 individ. m^{-2}) was contrasted with the control (n = 4 for each treatment).

Experiment 5

Experiment 5 was the only experiment using blue mussels, *Mytilus edulis*, and was carried out for 14 days from 26 September to 10 October. The control treatment was contrasted with two experimental treatments of 16 and 64 mussels per mesocosm for densities of 57 and 229 individ. m^{-2}, respectively (n = 4 for each treatment). These densities are commonly found in some areas of Long Island, but are much lower than those found in dense mussel beds in nearby Narragansett Bay, Rhode Island (814 – 9943 individ. m^{-2}; Altieri and Witman, 2006).

Statistical analysis

To compare differences in seagrass productivity between treatments, one-way ANOVA's and Tukey multiple comparison tests were carried out using the program SigmaStat 3.0. The dry weight of epiphytes on the seagrass was normalized by the area of the seagrass leaves, and also analyzed by treatment for each experiment using a one-way ANOVA. Since experiments 1 and 4 had only two treatments, *t*-tests were used in place of one-way ANOVAs. Chlorophyll *a* concentrations and light attenuation were

analyzed for each experiment using a two-way repeated measures ANOVA, with treatment and day as factors. A linear regression was used to examine correlations between leaf area production and light attenuation. All values in text are reported as mean ± SE.

RESULTS

Experiment 1

The temperature for all mesocosms in experiment 1 was 19.1°C ± 0.6 and the salinity was 21.96 ± 0.09. There was a remarkable difference in chl *a* concentrations over the course of the experiment between the control, which was 34.08 ± 9.54 μg L^{-1}, and the "clams" treatment, which was 3.04 ± 0.38 μg L^{-1} (Fig 1A). Through the experiment, the percentage of phytoplankton biomass in the >5 μm size fraction, as measured by chl *a*, was 33.4 ± 4.0% in the control treatment and 38.3 ± 6.7% in the "clams" treatment. The control treatment had greater light attenuation (1.101 ± 0.139 m^{-1}) than the "clams" treatment (0.814 ± 0.190 m^{-1}; Fig 1B). For this experiment, chl *a* concentrations and light attenuation varied significantly by treatment (Two-way RM ANOVA, $p<0.001$ for chl *a* and $p<0.01$ for light attenuation) and by day ($p<0.001$ for both). Concurrently, the leaf area productivity of the eelgrass in the control treatment (0.318 ± 0.02 cm^2 $shoot^{-1}$ d^{-1}) was significantly lower than eelgrass productivity in the the "clams" treatment (0.462 ± 0.04 cm^2 $shoot^{-1}$ d^{-1}; *t*-test, $p<0.05$; Fig 1C). The estimated turnover time of the water in the "clams" treatment was 1.1 days (Table 1).

Experiment 2

The average temperature for all mesocosms in experiment 2 was 20.3°C ± 0.6 and the average salinity was 26.29 ± 0.05. From day 2 to day 9, chl *a* values were consistently lowest in the high density clam treatment (24.31 ± 7.67 μg L^{-1}), intermediate in the low density clam treatment (33.85 ± 9.85 μg L^{-1}), and highest in the control (45.22 ± 14.33 μg L^{-1}; Fig 2A). The percentage of phytoplankton biomass in the >5 μm size fraction, as measured by chl *a*, was 32.2 ± 2.8% in the control treatment, 35.2 ± 3.5% in the low density treatment, and 37.6 ± 4.0% in the high density treatment. Light attenuation showed a similar pattern through day 8, with K_d being lowest in the high density treatment (0.648 ± 0.158 m^{-1}), intermediate in the low density treatment (0.813 ± 0.160 m^{-1}), and highest in the control (0.960 ± 0.203 m^{-1}; Fig 2B). Accordingly, chl *a* concentrations and light attenuation varied significantly by treatment and by day (Two-way RM ANOVA, $p<0.001$ in all cases). The low clam density (14 individ. m^{-2}) treatment produced a leaf area productivity (0.832 ± 0.04 cm^2 $shoot^{-1}$ d^{-1}), similar to that of the high density (29 individ. m^{-2}) treatment (0.806 ± 0.04 cm^2 $shoot^{-1}$ d^{-1}), but the eelgrass shoots in both clam treatments were significantly more productive than the control treatment (0.642 ± 0.07 cm^2 $shoot^{-1}$ d^{-1} (Fig 2C; One-way ANOVA, $p<0.05$). The estimated turnover time was 2.2 days for the high density treatment and 4.5 days for the low density treatment (Table 1).

Experiment 3

The average temperature for all mesocosms in experiment 3 was 23.2°C ± 0.5 and the average salinity was 24.23 ± 0.56. As in experiment 2, a clear gradation in chl *a* levels was observed between the high density (11.05 ± 4.18 μg L^{-1}) oyster treatment, the low density (15.75 ± 7.40 μg L^{-1}) oyster treatment, and the control (25.64 ± 9.78 μg L^{-1};

Fig 3A). The percentage of phytoplankton biomass in the >5 μm size fraction, as measured by chl *a*, was 38.0 ± 1.7% in the control treatment, 35.2 ± 3.5% in the low density treatment, and 34.0 ± 2.5% in the high density treatment. Light attenuation had similarly consistent pattern over days 1-5; K_d was lowest in the high density treatment (1.502 ± 0.268 m^{-1}), intermediate in the low density treatment (1.586 ± 0.229 m^{-1}), and highest in the control (1.726 ± 0.224 m^{-1}; Fig 3B). Chlorophyll *a* concentrations varied significantly by treatment and by day (Two-way RM ANOVA, $p<0.001$ in both cases); light attenuation varied significantly by treatment over days 1-5 (Two-way RM ANOVA, $p<0.05$) and significantly by day (Two-way RM ANOVA, $p<0.001$). Leaf area productivity was significantly higher in both the low density (0.495 ± 0.03 cm^2 $shoot^{-1}$ d^{-1}) and high density (0.548 ± 0.02 cm^2 $shoot^{-1}$ d^{-1}) treatments than in the control treatment (0.371 ± 0.03 cm^2 $shoot^{-1}$ d^{-1}; Tukey test, $p<0.05$ and $p<0.01$, respectively; Fig 3C). The estimated turnover times were 0.6 for the high density of oysters and 1.3 days for the low density treatment (Table 1).

Experiment 4

The average temperature for all mesocosms in experiment 4 was 24.6°C ± 0.2 and the average salinity was 21.37 ± 0.09. Experiment 4 produced a clear difference between the control and oyster treatment chl *a* concentrations that was mirrored by changes in algal biomass and light levels. The chl *a* in the control (49.16 ± 7.36 μg L^{-1}) was higher than in the oyster treatment (31.82 ± 4.57 μg L^{-1}; Fig 4A) while light attenuation was also higher in the control (1.688 ± 0.094 m^{-1}) than in the oyster treatment (1.239 ± 0.131 m^{-1}; Fig 4B). The percentage of phytoplankton biomass in the >5 μm size fraction, as measured by chl *a*, was 34.3 ± 4.7% in the control treatment and 28.4 ± 4.4% in the

oyster treatment. Chlorophyll *a* concentrations varied significantly by treatment and by day (Two-way RM ANOVA, $p<0.05$ and $p<0.01$, respectively). Light attenuation also varied between treatments and by day (Two-way RM ANOVA, $p<0.05$ and $p<0.001$, respectively). Leaf area productivity was significantly higher in the experimental treatments with one oyster (0.560 ± 0.02 cm^2 shoot^{-1} d^{-1}) than in the control treatments (0.355 ± 0.04 cm^2 shoot^{-1} d^{-1}; two-tailed *t*-test, $p<0.01$; Fig 4C). The turnover time for the oyster treatment was 2.5 days (Table 1).

Experiment 5

The average temperature for all mesocosms in experiment 5 was $19.3°C \pm 0.7$ and the average salinity was 28.88 ± 0.09. Unlike experiments 2 and 3, there was not a consistent relationship between mussel density and chl *a* levels; chl *a* was actually higher in the low density treatment than in the control treatment on day 7. The high density mussel treatment, however, had consistently lower chl *a* (15.45 ± 5.40 μg L^{-1}) than the low density (40.11 ± 8.16 μg L^{-1}) and the control (40.37 ± 7.43 μg L^{-1}; Fig 5A). The percentage of phytoplankton biomass in the >5 μm size fraction, as measured by chl *a*, was $24.8 \pm 5.2\%$ in the control treatment, $29.2 \pm 2.5\%$ in the low density treatment, and $14.2 \pm 5.4\%$ in the high density treatment. Chlorophyll *a* levels varied significantly by treatment and by day (Two-way RM ANOVA, $p<0.05$ and $p<0.001$, respectively). Unfortunately, we were not able to obtain light data for experiment 5, although the water in the high density mussel treatment was visibly clearer to than the low density treatment and the control. Leaf area productivity was 0.790 ± 0.11 cm^2 shoot^{-1} d^{-1} in the treatments with a high density (229 individ. m^{-2}) of mussels, 0.435 ± 0.02 cm^2 shoot^{-1} d^{-1} in the low density (57 individ. m^{-2}) treatment, and 0.399 ± 0.08 cm^2 shoot^{-1} d^{-1} in the control

treatment (Fig 5B). The productivity was significantly higher in the high density treatment but did not significantly differ between the low density and the control treatments (1-way ANOVA and Tukey multiple comparison, $p<0.05$). The estimated turnover times were 3.6 days for the high density of mussels and 14.5 days for the low density of mussels (Table 1).

Bivalve filtration rates, turnover times, and facilitation of eelgrass productivity

Length, weight, and clearance rate measurements were recorded for each group of bivalves (Table 1). In all cases, the bivalves filtered from the control treatment mesocosms at a significant rate (Table 1). *Crassostrea virginica*, which had the highest individual and weight-specific clearance rates (1.91 ± 0.97 L hr^{-1} g^{-1} AFDW), produced the shortest estimates for mesocosm turnover time (0.6 – 2.5 d; Table 1), followed by *M. mercenarina* (clearance rate = 0.41 ± 0.24 L hr^{-1} g^{-1} AFDW; turnover time = 1.1 – 4.5 d), and *M. edulis* (clearance rate = 0.29 ± 0.29 L hr^{-1} g^{-1} AFDW; turnover time = 3.6 – 14.5 d). Epibiont biomass, as measured by mg AFDW of epibionts cm^{-2} leaf area, did not differ significantly among treatments in any experiment (data not shown).

During our experiments, higher densities of bivalves produced dramatic decreases in water column chl *a* over the course of each experiment (Fig 1A,2A,3A,4A,5A). Experiments 1-4 also had significant decreases in light attenuation in the treatments with bivalves (Fig 1B,2B,3B,4B). There was a significant inverse correlation (Fig 6; r^2 = 0.400, $p<0.001$) between leaf area productivity and mean light attenuation coefficient for experiments 1-4 (light attenuation data was not available for experiment 5). There was also a significant inverse correlation between mesocosm turnover time (Table 1) and leaf

area productivity among all experiments ($y = -0.015x + 0.647$, $r^2 = 0.21$, $p<0.05$; regression not shown).

DISCUSSION

This study has demonstrated, through a series of mesocosm experiments, that suspension-feeding bivalves can facilitate the growth of eelgrass. Over the course of five experiments, the effects of three densities of *Mercenaria mercenaria*, three densities of *Crassostera virginica*, and two densities of *Mytilus edulis* were examined. In all cases, the highest density of bivalves produced significant decreases in chl *a*, increases in light penetration, and significant increases in leaf area productivity of *Zostera marina*. On average, eelgrass growth increased by of 48 ± 9.3% in the presence of moderate densities of bivalves relative to control treatments. For *M. mercenaria* and *C. virginica*, intermediate or even low densities of these species filtered sufficiently to alter light and chl *a* levels to the benefit of eelgrass productivity. The results of these experiments help to refine our understanding of the function of filter-feeding bivalves in estuarine ecosystems.

Some studies have suggested that suspension-feeding bivalves control eutrophication and algal blooms (Officer et al., 1982; Cerrato et al., 2004; Cloern, 1982) or significantly alter carbon-cycling (Doering et al., 1986). Newell and Koch's (2004) modeling study predicted that filtration by bivalves could benefit seagrass. To our knowledge, this is the first study that demonstrates the facilitation of eelgrass growth by the filtration of bivalves in an experimental setting. The mechanism of facilitation is an increase in light penetration (Fig 1B,2B,3B,4B), paired with dramatic reductions in the

standing stocks of phytoplankton (Fig 1A,2A,3A,4A,5A), due to the bivalves' clearance of the water column. Other studies (Reusch et al., 1994; Peterson and Heck, 2001) have demonstrated that nutrient fertilization by bivalves through biodeposition can enhance growth of seagrass. Peterson and Heck's (2001) study was carried out in St. Joseph Bay, Florida, an oligotrophic environment where light was plentiful and nutrients were scarce. Because of the eutrophic nature of a great number of estuaries (Nixon, 1995; de Jonge et al., 2002; Kemp et al., 2005; Valiela, 2006), mitigation of light limitation may be an even more common mechanism by which bivalve filtration benefits seagrass populations. This study was not designed to separate the relative contributions of nutrient fertilization and water transparency effects on seagrasses by bivalves in eutrophic estuaries.

During our experiments, higher densities of bivalves produced dramatic decreases in water column chl *a* and light attenuation (Fig 1,2,3,4,5) and there was a significant inverse correlation (Fig 6; $r^2 = 0.400$, $p<0.001$) between leaf area productivity and mean light attenuation coefficient. The decreases in chl *a*, increases in light penetration, and correlation between leaf area productivity and light levels suggest that the principle effect of the bivalves on eelgrass growth was mediated by clearing of the water column leading to increased light penetration. During our experiments, chlorophyll *a* levels tended to decrease ($\leq$20 μg L^{-1}) toward the end of each experiment in all treatments (Fig 1A,2A,3A,4A,5A), likely due to the development of high levels of algal biomass (> 60 μg L^{-1}) in control tanks whose nutrient demand greatly exceeded our nutrient loading rate (10 μM ammonium and 0.625 μM orthophosphate daily). Had our nutrient loading rate increased concurrently with increasing algal biomass to sustain the high biomass levels throughout the experiment, the significant differences in seagrass productivity between

control and shellfish treatments would have likely been even larger than observed (Fig 1C,2C,3C,4C,5B).

The depth of the water column in our experiments was 1.2 m, a depth comparable to some northeast US lagoons such as Great South Bay, Waquoit Bay (Valiela et al., 1992), and Barnegat Bay (Bologna et al., 2005) or European estuaries such as the Wadden Sea (Smaal and Prins, 1993), but shallower than systems such as Chesapeake Bay (Kemp et al., 2005), San Francisco Bay (Officer et al., 1982), or the Baltic Sea (Smaal and Prins, 1993). Smaal and Prins (1993) surveyed bivalve suspension feeding in several European estuaries and defined "filtration pressure" as the ratio of bivalve consumption to phytoplankton production in the overlying water column. Obviously, as the water column depth increases, the density of benthic suspension feeders required to balance the production in the overlying water column also increases. Smaal and Prins (1993) suggested that a density of 2-8 g AFDW bivalve tissue m^{-3} of water column was enough for bivalve suspension feeders to exert a strong influence on the overlying water column. Our experimental bivalve densities (7.5-79.1 g AFDW m^{-3}) met or exceeded this range of biomass, indicating that all of our experimental bivalve densities should have been able to clear the volume of the mesocosms. The lowest density of mussels, which did not exert a significant influence on chl *a*, light, or eelgrass growth, had a biomass of 9.1 g AFDW m^{-3}, above Smaal and Prins' (1993) mass requirement. This suggests that the individual or weight-specific clearance rate of a given suspension feeder may be more important than the total biomass or that lower temperatures present during this final experiment contributed lower filtration rates.

Some recent studies have focused on the turnover time, or clearance time, for suspension feeders to filter the volume of a body of water (Cerrato et al., 2004; Newell and Koch, 2004; Bologna et al., 2005). In a previous mesocosm experiment with *Mercenaria mercenaria*, Cerrato et al. (2004) found that clearance times of 0.51 to 2.4 days were sufficient to prevent the development of dense brown tide blooms, while blooms proliferated at clearance times of 3.7 days or longer. Our clearance times, based upon the measured clearance rates of bivalves used in our experiments, ranged from 0.64 days for our highest density of oysters to 14.5 days for our lowest density of mussels (Table 1). Interestingly, at the longest clearance time of 14.5 days, chl *a* levels and eelgrass growth in the low-density mussel treatment was not significantly different from the control. The clearance time (4.5 days) for the lowest density of hard clams, while longer than the critical values in Cerrato et al.'s (2004) study, did produce a significant decrease in chl *a* and a significant increase in eelgrass growth. All other clearance times were $\leq$ 3.6 days and also produced significant increases in eelgrass growth. Since these clearance times are based on clearance measurements for bivalves placed in water with high algal biomass (>20 $\mu g\ l^{-1}$ chl *a*), clearance rates for bivalves feeding at lower concentrations of chl *a* may have been higher (Clausen and Riisgard, 1996).

The results of these mesocosm experiments, combined with the work of Officer et al. (1982), Cloern (1982), Cerrato et al. (2004), Newell and Koch (2004), and many others, suggest dense communities of benthic suspension feeders can serve as a control on the negative effects of eutrophication. Clearly, this benefits seagrass productivity through increased light penetration (Fig 1C,2C,3C,4C,5B). There are likely many other synergistic interactions between bivalves and seagrasses which facilitate growth and

recruitment of both clades, perhaps to the benefit of entire ecosystems. Bivalves clear the water column and increase light penetration for seagrasses and benthic diatoms (Lotze et al., 2006), while seagrasses provide habitat, predation refuges, and a benthic source of oxygen for bivalves and other organisms (Valiela et al., 1992; Reise, 2002; Bruno et al., 2003; Bologna et al., 2005). Bivalves also fertilize seagrass roots through biodeposition (Peterson and Heck, 1999, 2001). Seagrasses can minimize benthic nutrient fluxes to the water column by stabilizing sediments and absorbing benthic nutrients (Reise, 2002; Bruno et al., 2003). These effects can work, together with bivalve filtration, to reduce suspended sediment load and minimize pelagic phytoplankton abundances (Newell and Koch, 2004; Lotze et al., 2006).

Recently, Pomeroy et al (2006) have suggested that oyster restoration would be unlikely to counter the effects of eutrophication in Chesapeake Bay, MD, USA, due to temporal and spatial decoupling of bivalve filtration pressure and algal blooms in this system. While they present several valid arguments, these points do not apply to the current study for the following reasons. Our mesocosms were meant to mimic a shallow, lagoon-type estuary, such as Long Island's (NY, USA) south shore estuaries, which have a mean depth of 1.2 m (Wilson et al 1991). In these systems, the water column is chronically well-mixed (Wilson et al 1991) and bivalves are evenly distributed (Weiss et al 2007), suggesting bivalves, algal blooms, and seagrasses should be more spatially coupled than in Chesapeake Bay. In addition, eelgrass growth and maximum bivalve filtration rates coincide during late spring through fall months (Grizzle et al 2001, Hemminga and Duarte 2001), providing a close temporal link between bivalves and eelgrass in temperate lagoonal systems.

A productive, high-biomass benthic community seems to be one of the hallmarks of a healthy estuary, and many anthropogenic insults drive estuaries into phytoplankton- and microbial-dominated systems at the expense of the benthic community (Jackson, 2001; Kemp et al., 2005; Lotze et al., 2006). These experiments indicate that a healthy benthos (robust bivalves and seagrass populations) is more resistant to eutrophication than seagrass alone. Estuaries become more vulnerable to eutrophication, algal blooms, hypoxia, and degradation of benthic habitats when overharvesting or habitat loss removes the filtration pressure of bivalves. For example, Cerco and Noel's (2007) modeling study predicted a tenfold increase in oyster biomass would lead to decreases in phytoplankton biomass and benthic nutrient fluxes and increases in dissolved oxygen and submerged aquatic vegetation (SAV).

The bivalve densities used in our mesocosms (14-57 clams m^{-2} and 4-14 oysters m^{-2}) were higher than current densities in many US estuaries such as Great South Bay or Chesapeake Bay, but were lower than historical densities found in these same systems (53-105 clams m^{-2} and 43-150 oysters m^{-2}, respectively; Cerrato et al., 2004; Newell and Koch, 2004). Jackson (2001) and Lotze et al. (2006) surveyed historical declines across a broad suite of organisms and habitats, and found that overharvesting and habitat destruction preceded eutrophication in most estuaries. As such, it would seem successful management efforts will need to take a 'ecosystem-based' approach which incorporates habitat conservation, shellfish restoration, and restrictions on nutrient loading to restore healthy estuarine function. In light of the results from this study, it would seem that even a partial recovery of shellfish populations could help combat eutrophication and have a beneficial impact on seagrass habitats in shallow, eutrophied estuaries.

References:

Altieri AH, Witman JD (2006) Local extinction of a foundation species in a hypoxic estuary: Integrating individuals to ecosystem. Ecology 87:717-730

Bintz JC, Nixon SW (2001) Responses of eelgrass Zostera marina seedlings to reduced light. Marine Ecology-Progress Series 223:133-141

Bintz JC, Nixon SW, Buckley BA, Granger SL (2003) Impacts of temperature and nutrients on coastal lagoon plant communities. Estuaries 26:765-776

Bologna PAX, Fetzer ML, McDonnell S, Moody EM (2005) Assessing the potential benthic-pelagic coupling in episodic blue mussel (Mytilus edulis) settlement events within eelgrass (Zostera marina) communities. Journal of Experimental Marine Biology and Ecology 316:117-131

Bruno JF, Stachowicz JJ, Bertness MD (2003) Inclusion of facilitation into ecological theory. Trends in Ecology & Evolution 18:119-125

Brush MJ, Nixon SW (2002) Direct measurements of light attenuation by epiphytes on eelgrass Zostera marina. Marine Ecology-Progress Series 238:73-79

Cerco CF, Noel MR (2007) Can oyster restoration reverse cultural eutrophication in Chesapeake Bay? Estuaries and Coasts 30:331-343

Cerrato RM, Caron DA, Lonsdale DJ, Rose JM, Schaffner RA (2004) Effect of the northern quahog Mercenaria mercenaria on the development of blooms of the brown tide alga Aureococcus anophagefferens. Marine Ecology-Progress Series 281:93-108

Clausen I, Riisgard HU (1996) Growth, filtration and respiration in the mussel Mytilus edulis: No evidence for physiological regulation of the filter-pump to nutritional needs. Marine Ecology-Progress Series 141:37-45

Cloern JE (1982) Does the Benthos Control Phytoplankton Biomass in South-San-Francisco Bay. Marine Ecology-Progress Series 9:191-202

de Jonge VN, Elliott M, Orive E (2002) Causes, historical development, effects and future challenges of a common environmental problem: eutrophication. Hydrobiologia 475:1-19

Dennison WC, Marshall GJ, Wigand, C (1989) Effects of "brown tide" shading on eelgrass (*Zostera marina*) distributions. In: Cosper EM, Bricelj VM, Carpenter EJ (eds) Novel phytoplankton blooms: causes and impacts of recurrent brown tides and other unusual blooms. Springer-Verlag, Berlin, p 675-692

Dennison WC, Alberte RS (1985) Role of Daily Light Period in the Depth Distribution of Zostera-Marina (Eelgrass). Marine Ecology-Progress Series 25:51-61

Doering PH, Oviatt CA, Kelly JR (1986) The Effects of the Filter-Feeding Clam Mercenaria-Mercenaria on Carbon Cycling in Experimental Marine Mesocosms. Journal of Marine Research 44:839-861

Duarte CM (1995) Submerged aquatic vegetation in relation to different nutrient regimes. Ophelia 41: 87-112

Gobler CJ, Boneillo GE (2003) Impacts of anthropogenically influenced groundwater seepage on water chemistry and phytoplankton dynamics within a coastal marine system. Marine Ecology-Progress Series 255:101-114

Gobler CJ, Lonsdale DJ, Boyer GL (2005) A review of the causes, effects, and potential management of harmful brown tide blooms caused by Aureococcus anophagefferens (Hargraves et Sieburth). Estuaries 28:726-749

Grizzle RE, Bricelj VM, Shumway SE (2001) Physiological ecology of *Mercenaria mercenaria* In: Biology of the Hard Clam, J.N. Kraeuter and M. Castagna, (Eds.), Elsevier Science, Amsterdam, pp. 305- 82.

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

Heath RT, Fahnenstiel GL, Gardner WS, Cavaletto JF, Hwang SJ (1995) Ecosystem-level effects of zebra mussels (Dreissena polymorpha): An enclosure experiment in Saginaw Bay, Lake Huron. Journal of Great Lakes Research 21:501-516

Hemminga MA, Duarte CM (2001) Seagrass Ecology. Cambridge University Press, Cambridge, UK, 310 pp

Heck KL, Wetstone GS (1977) Habitat Complexity and Invertebrate Species Richness and Abundance in Tropical Seagrass Meadows. Journal of Biogeography 4:135-142

Jackson JBC (2001) What was natural in the coastal oceans? Proceedings of the National Academy of Sciences of the United States of America 98:5411-5418

Kemp WM, Boynton WR, Adolf JE, Boesch DF, Boicourt WC, Brush G, Cornwell JC, Fisher TR, Glibert PM, Hagy JD, Harding LW, Houde ED, Kimmel DG, Miller WD, Newell RIE, Roman MR, Smith EM, Stevenson JC (2005) Eutrophication of Chesapeake Bay: historical trends and ecological interactions. Marine Ecology-Progress Series 303:1-29

Lotze HK, Lenihan HS, Bourque BJ, Bradbury RH, Cooke RG, Kay MC, Kidwell SM, Kirby MX, Peterson CH, Jackson JBC (2006) Depletion, degradation, and recovery potential of estuaries and coastal seas. Science 312:1806-1809

Newell RIE, Koch EW (2004) Modeling seagrass density and distribution in response to changes in turbidity stemming from bivalve filtration and seagrass sediment stabilization. Estuaries 27:793-806

Nixon SW (1995) Coastal marine eutrophication – A definition, social causes, and future concerns. Ophelia 41:199-219

Officer CB, Smayda TJ, Mann R (1982) Benthic Filter Feeding - a Natural Eutrophication Control. Marine Ecology-Progress Series 9:203-210

Parsons TR, Maita Y, Lalli CM (1984) A manual of chemical and biological methods for seawater analysis. Pergamon Press, Oxford

Peterson BJ, Heck KL (1999) The potential for suspension feeding bivalves to increase seagrass productivity. Journal of Experimental Marine Biology and Ecology 240:37-52

Peterson BJ, Heck KL (2001) An experimental test of the mechanism by which suspension feeding bivalves elevate seagrass productivity. Marine Ecology-Progress Series 218:115-125

Pomeroy LR, D'Elia CF, Schaffner LC (2006) Limits to top-down control of phytoplankton by oysters in Chesapeake Bay. Mar Ecol Prog Ser 325:301-309

Reise K (2002) Sediment mediated species interactions in coastal waters. Journal of Sea Research 48:127-141
Reusch TBH, Chapman ARO, Groger JP (1994) Blue Mussels Mytilus-Edulis Do Not Interfere with Eelgrass Zostera-Marina but Fertilize Shoot Growth through Biodeposition. Marine Ecology-Progress Series 108:265-282
Riisgard HU (2001) On measurement of filtration rates in bivalves - the stony road to reliable data: review and interpretation. Marine Ecology-Progress Series 211:275-291
Smaal AC, Prins TC (1993) The uptake of organic matter and the release of inorganic nutrients by bivalve suspension feeder beds. In: Dame RF (ed) Bivalve filter feeders in estuarine and coastal ecosystem processes. Springer-Verlag, Berlin, p 271-298
Sokal RR, Rohlf FJ (1995) Biometry. WH Freeman and Co., New York, 887 pp
Taylor DI, Nixon SW, Granger SL, Buckley BA (1999) Responses of coastal lagoon plant communities to levels of nutrient enrichment: A mesocosm study. Estuaries 22:1041-1056
Valiela I (2006) Global coastal change. Blackwell Publishing, Boston, 376 pp
Valiela I, Foreman K, Lamontagne M, Hersh D, Costa J, Peckol P, Demeoandreson B, Davanzo C, Babione M, Sham CH, Brawley J, Lajtha K (1992) Couplings of Watersheds and Coastal Waters - Sources and Consequences of Nutrient Enrichment in Waquoit Bay, Massachusetts. Estuaries 15:443-457
Weiss MB, Curran PB, Peterson BJ, Gobler CJ (2007) The influence of plankton composition and water quality on hard clam (*Mercenaria mercenaria* L.) populations across Long Island's south shore lagoon estuaries. J Exp Mar Biol Ecol 345:12–25
Wilson RE, Wong KC, Carter HH (1991) Aspects of circulation and exchange in Great South Bay, p.33-42. In: J.R. Schubel, T.M. Bell and H.H. Carter [eds.] The Great South Bay. SUNY press, Albany, NY.
Zieman JC (1974) Methods for Study of Growth and Production of Turtle Grass, Thalassia-Testudinum Konig. Aquaculture 4:139-143

Table 1. Mean sizes, weights, and AFDW-normalized clearance rates for the three bivalve species. Measurements were taken once per species, and mean individual clearance rates were used to estimate turnover times. All measures are mean ± SE.

Bivalve species	Size (longest shell dimension, mm)	Ash-free dry tissue weight (g AFDW)	Clearance rate measured (L h^{-1} g^{-1} AFDW)	Estimated Turnover Time (days)
Hard Clam, (Quahog) *Mercenaria mercenaria*	52.2 ± 0.5	1.688 ± 0.158	0.41 ± 0.03	14 Clams m^{-2}: 4.5d 29 Clams m^{-2}: 2.2 d 57 Clams m^{-2}: 1.1 d
Eastern Oyster, *Crassostrea virginica*	84.5 ± 1.1	2.564 ± 0.149	1.91 ± 0.28	4 Oysters m^{-2}: 2.5 d 7 Oysters m^{-2}: 1.3 d 14 Oysters m^{-2}: 0.6 d
Blue Mussel, *Mytilus edulis*	39.0 ± 0.7	0.195 ± 0.020	0.289 ± 0.097	57 Mussels m^{-2}: 14.5 d 229 Mussels m^{-2}: 3.6 d

Figure Legends

Figure 1. Temporal changes in (A) chlorophyll *a* (chl a μg L^{-1}) and (B) extinction coefficient (K_d m^{-1}) for experiment 1. C) Differences in eelgrass leaf area production. Clams are *Mercenaria mercenaria*. All error bars are ± SE.

Figure 2. Temporal changes in (A) chlorophyll *a* (chl a μg L^{-1}) and (B) extinction coefficient (K_d m^{-1}) for experiment 2. C) Differences in eelgrass leaf area production. Clams are *Mercenaria mercenaria*. All error bars are ± SE.

Figure 3. Temporal changes in (A) chlorophyll *a* (chl a μg L^{-1}) and (B) extinction coefficient (K_d m^{-1}) for experiment 3. C) Differences in eelgrass leaf area production. Oysters are *Crassostrea virginica*. All error bars are ± SE.

Figure 4. Temporal changes in (A) chlorophyll *a* (chl a μg L^{-1}) and (B) extinction coefficient (K_d m^{-1}) for experiment 4. C) Differences in eelgrass leaf area production. Oysters are *Crassostrea virginica*. All error bars are ± SE.

Figure 5. Temporal changes in (A) chlorophyll *a* (chl a μg L^{-1}) for experiment 5. B) Differences in eelgrass leaf area production. Mussels are *Mytilus edulis*. All error bars are ± SE.

Figure 6. Changes in leaf area productivity as a function of mean K_d for all tanks from experiments 1-4. Symbols are as follows: ▲, experiment 1; Δ, experiment 2; ●, experiment 3; ○, experiment 4.

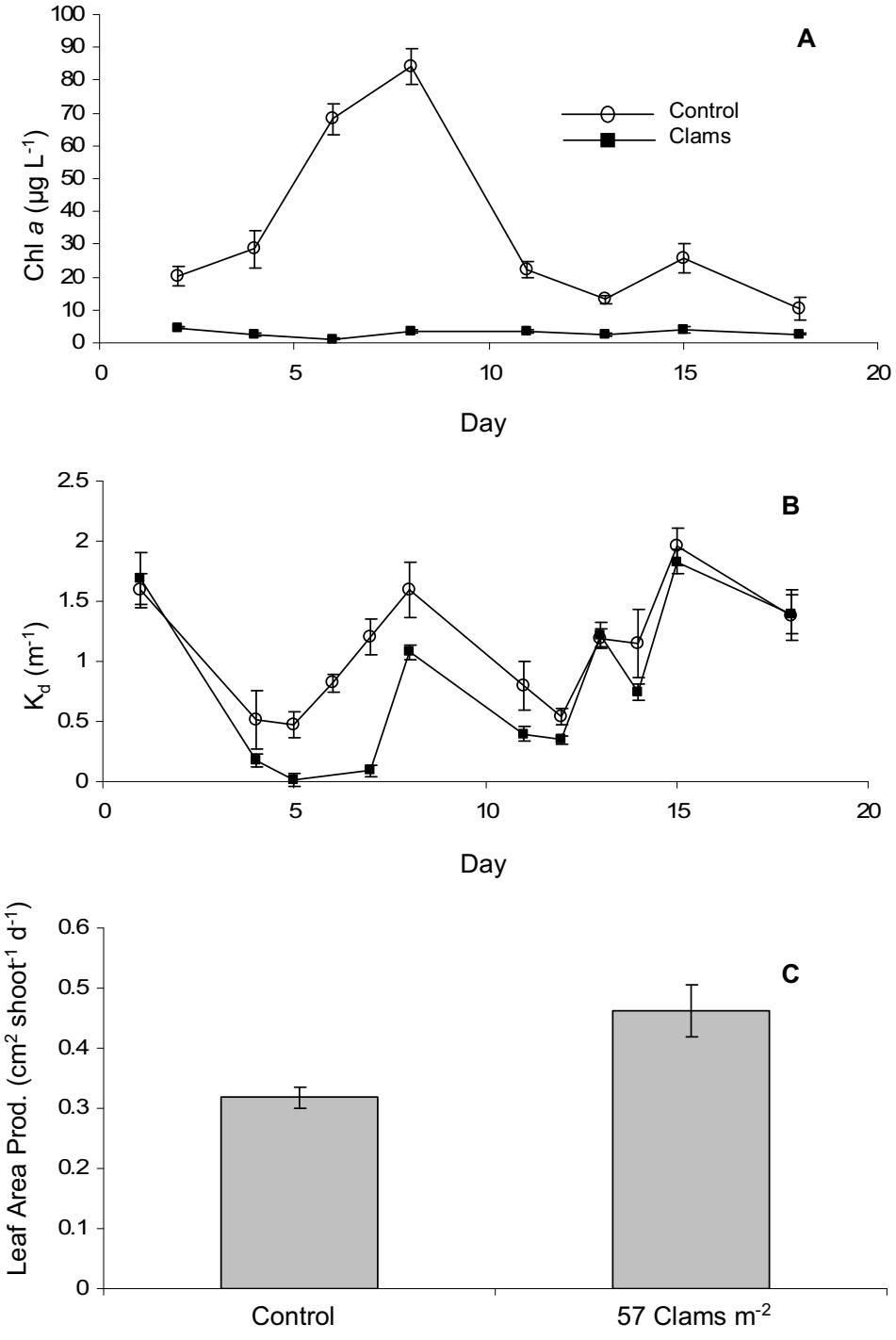

A
Control
Clams
Chl a (µg L-1)
Day
B
Kd (m-1)
Day
C
Leaf Area Prod. (cm2 shoot-1 d-1)
Control
57 Clams m-2

Figure 1

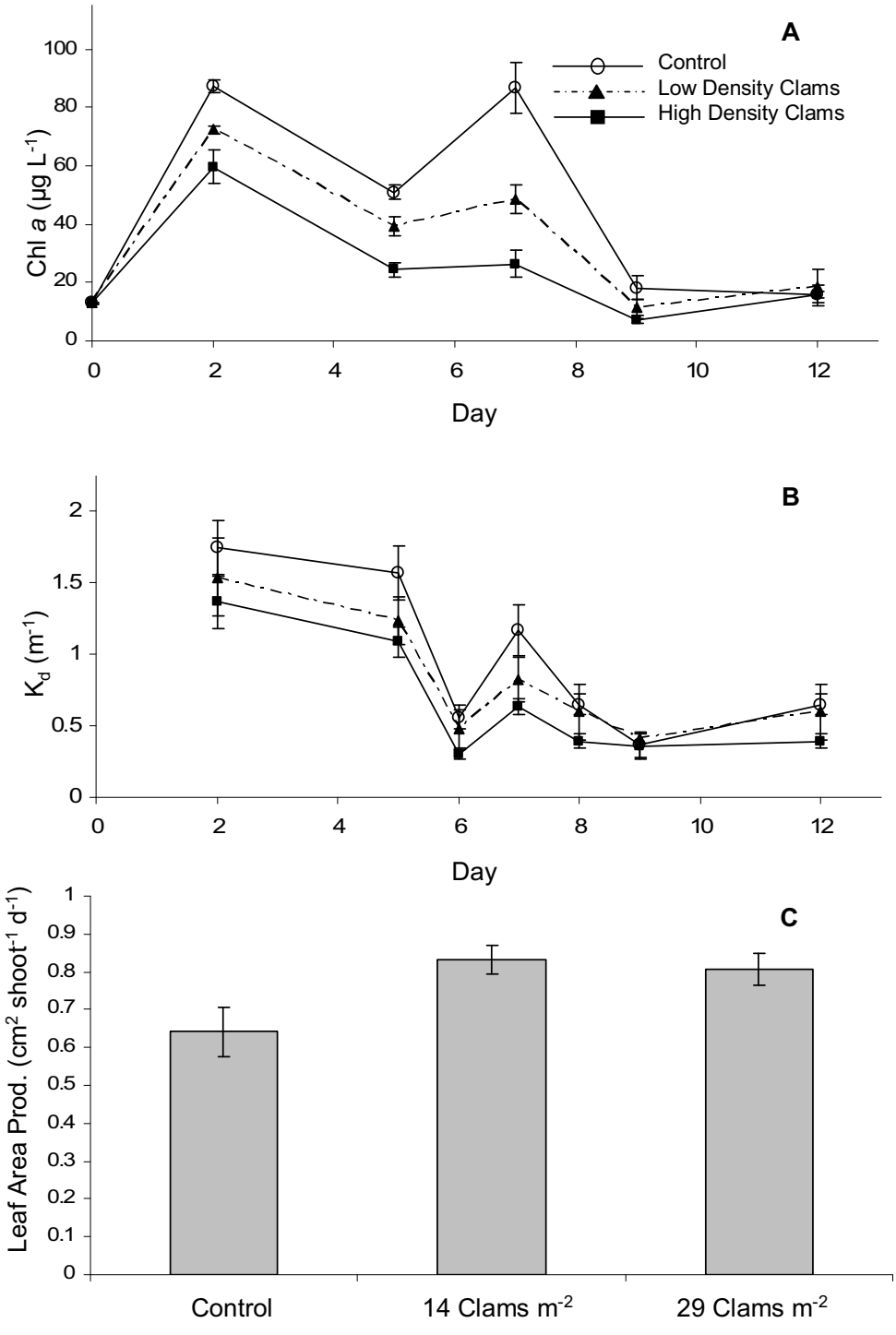

A
Control
Low Density Clams
High Density Clams
Chl a (µg L-1)
Day
B
Kd (m-1)
Day
C
Leaf Area Prod. (cm2 shoot-1 d-1)
Control
14 Clams m-2
29 Clams m-2

Figure 2

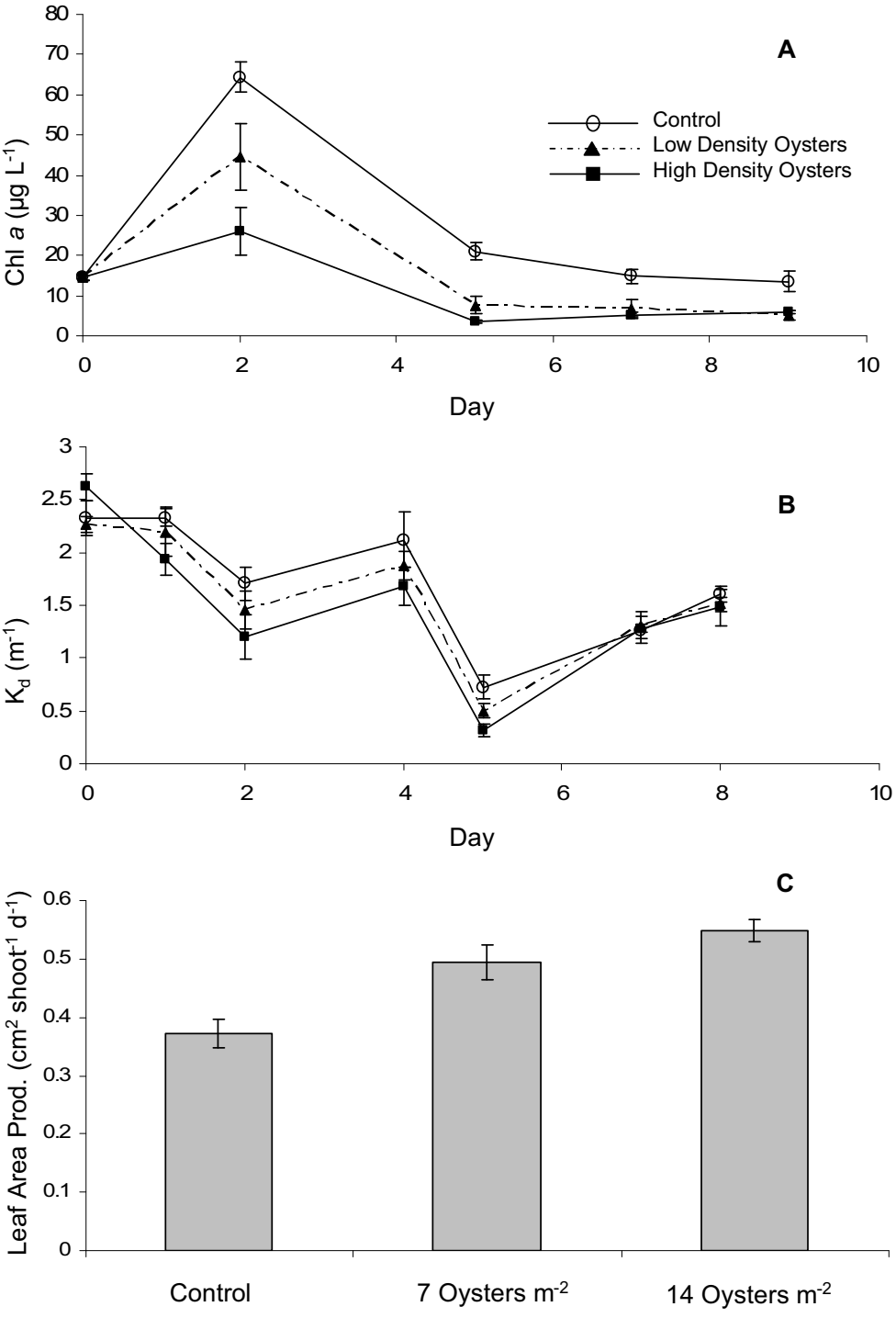

A
Control
Low Density Oysters
High Density Oysters
Chl a (µg L-1)
Day
B
Kd (m-1)
Day
C
Leaf Area Prod. (cm2 shoot-1 d-1)
Control
7 Oysters m-2
14 Oysters m-2

Figure 3

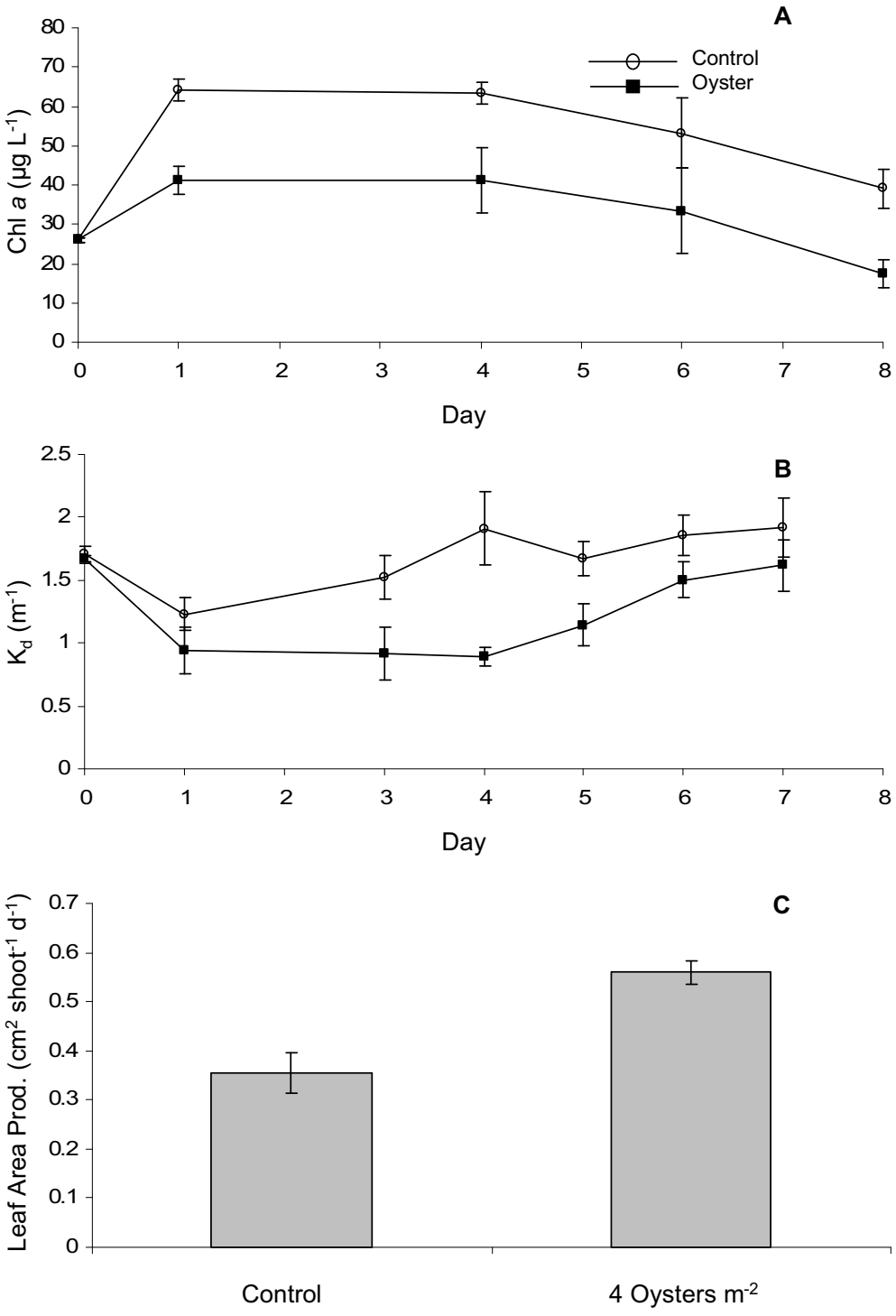

A
Control
Oyster
Chl a (µg L-1)
Day
B
Kd (m-1)
Day
C
Leaf Area Prod. (cm2 shoot-1 d-1)
Control
4 Oysters m-2

Figure 4

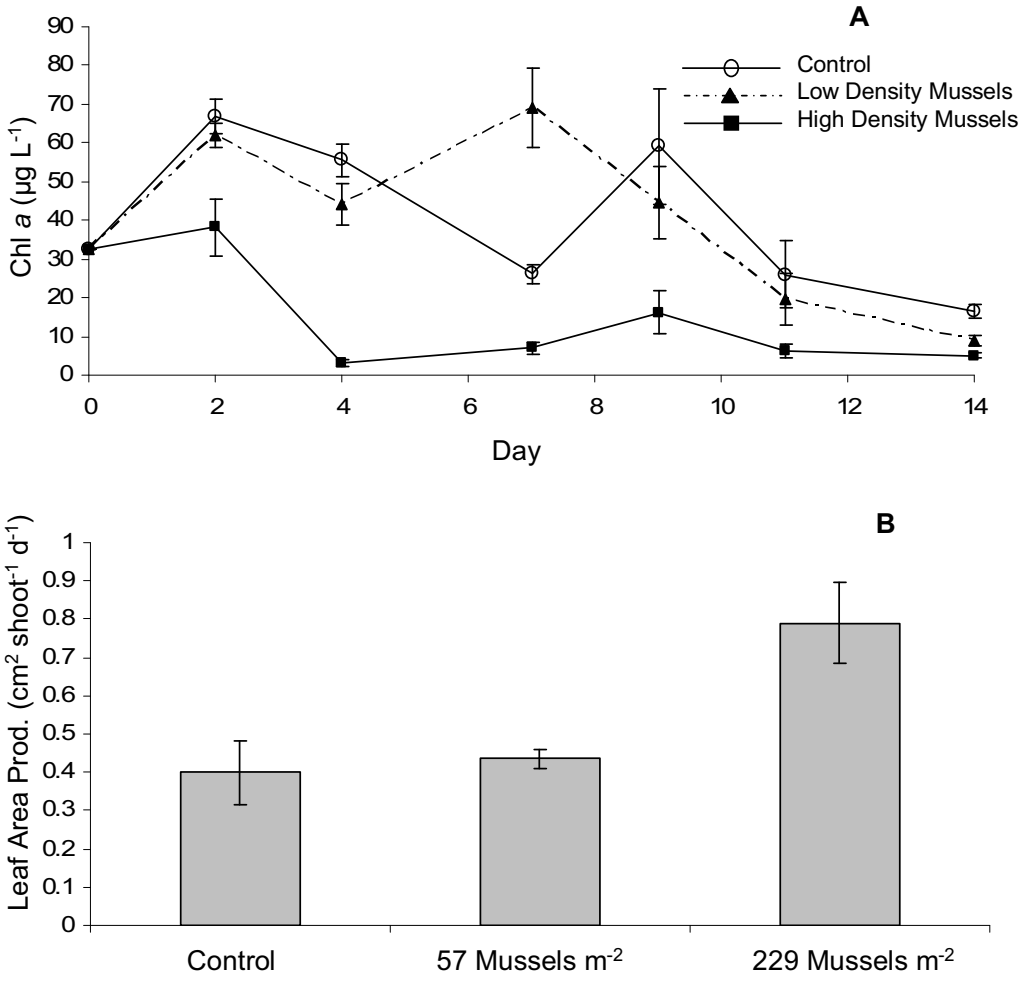

A
Control
Low Density Mussels
High Density Mussels
Chl a (µg L-1)
Day
B
Leaf Area Prod. (cm2 shoot-1 d-1)
Control
57 Mussels m-2
229 Mussels m-2

Figure 5

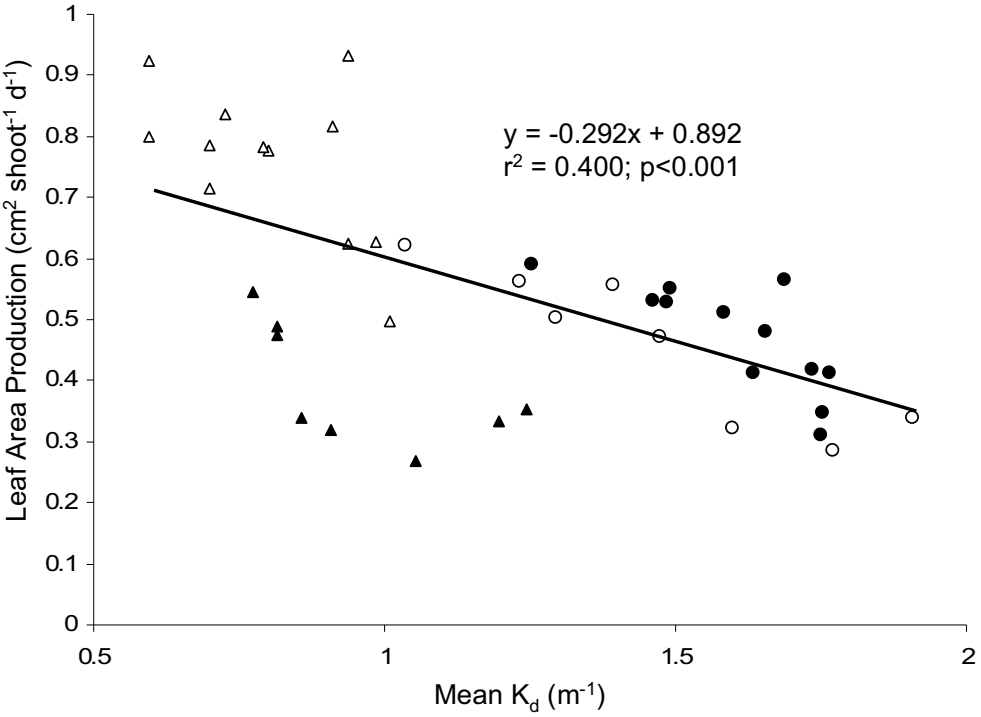

y = -0.292x + 0.892
r2 = 0.400; p<0.001
Leaf Area Production (cm2 shoot-1 d-1)
Mean Kd (m-1)
0
0.1
0.2
0.3
0.4
0.5
0.6
0.7
0.8
0.9
1
0.5
1
1.5
2

Figure 6