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The Effects of Multiple Stressors on the Balance between Autotrophic and Heterotrophic Processes in an Estuarine System

TRACY N. WIEGNER[1,*], SYBIL P. SEITZINGER[1], DENISE L. BREITBURG[2], and JAMES G. SANDERS[2,†]

[1] *Institute of Marine and Coastal Sciences, Rutgers, The State University of New Jersey, 71 Dudley Road, New Brunswick, New Jersey 08901-8521*

[2] *The Academy of Natural Sciences, Estuarine Research Center, 10545 Mackall Road, St. Leonard, Maryland 20685*

ABSTRACT: Responses of autotrophic and heterotrophic processes to nutrients and trace elements were examined in a series of experimental estuarine food webs of increasing trophic complexity using twenty 1-m^3 mesocosms. Nutrients (nitrogen and phosphorus) and trace elements (a mix of arsenic, copper, cadmium) were added alone and in combination during four experimental runs spanning from spring 1997 to spring 1998. Diel changes in dissolved oxygen were used to examine whole system gross primary production (WS-GPP), respiration (WS-RESP), and net ecosystem metabolism (NEM). Nutrient and trace element additions had the greatest effect on WS-GPP, WS-RESP, and NEM; trophic complexity did not significantly affect any of these parameters ($p > 0.3$). Effects of trophic complexity were detected in nutrient tanks where bivalves significantly ($p = 0.03$) reduced WS-GPP. Nutrient additions significantly enhanced WS-GPP and to a lesser extent WS-RESP during most mesocosm runs. The system shifted from net heterotrophy (-17.2 ± 1.8 mmol C m^{-3} d^{-1}) in the controls to net autotrophy (29.1 ± 7.6 mmol C m^{-3} d^{-1}) in the nutrient tanks. The addition of trace elements alone did not affect WS-GPP and WS-RESP to the same extent as nutrients, and their effects were more variable. Additions of trace elements alone consistently made the system more net heterotrophic (-24.9 ± 1.4 mmol C m^{-3} d^{-1}) than the controls. When trace elements were added in combination with nutrients, the nutrient-enriched system became less autotrophic (1.6 ± 3.1 mmol C m^{-3} d^{-1}). The effects of trace elements on NEM occurred primarily through reductions in WS-GPP rather than increases in WS-RESP. Our results suggest that autotrophic and heterotrophic processes respond differently to these stressors.

Introduction

Worldwide, coastal ecosystems are increasingly being overwhelmed by both natural and anthropogenic stressors. These stressors can be physical, chemical, or biological in nature and include factors as wide ranging as hurricanes, anthropogenic contaminants, and invasive species. Historically, aquatic research has focused on determining the impact of a single stressor on an ecosystem. A significant body of this research has centered on the impacts of increased nutrients to the coastal zone. While these studies have provided much insight into the functioning of estuaries, they have only begun to reveal how these systems respond to stressors. As more coastal environments become impacted by human activities, ascertaining the synergistic and antagonistic interaction among various stressors, as well as determining their effect on the ecosystem, will become essential (Breitburg et al. 1998).

Nutrients and trace element contaminants are two stressors affecting estuaries. Increased nutrient loads to estuaries have resulted in increased nuisance and toxic algal blooms, fish and shellfish kills, hypoxic and anoxic bottom waters, and habitat degradation (Rosenberg 1985; Howarth 1988; Jickells 1998). Concentrated trace element contaminant loads to estuaries are more localized to urban and industrial areas (Förstner 1983a; Sanders and Cibik 1988; Nriagu 1990). At high concentrations, these contaminants are toxic to both aquatic biota and humans (Förstner 1983b; Nriagu 1990). Persistent low-level inputs of trace elements from point and non-point sources are more widespread and are becoming recognized as a threat to estuaries (Sanders and Cibik 1988; Sañudo-Wilhelmy and Gill 1999). They have been shown to change plankton and benthic community structure and productivity (Beers et al. 1977; Gamble et al. 1977; Reeve et al. 1977; Sanders et al. 1981; Sanders and Cibik 1985a; Klerks and Levinton 1989). While a considerable amount is known about how nutrients or trace element contaminants alone impact estuaries, less is known about their combined effects.

One way to assess how a coastal ecosystem will

* Corresponding author; current address: Stroud Water Research Center, 970 Spencer Road, Avondale, Pennsylvania 19311; tele: 610/268-2153 ext. 257; fax: 610/268-0490; e-mail: twiegner@stroudcenter.org.

† Current address; Skidaway Institute of Oceanography, 10 Ocean Science Circle, Savannah, Georgia 31411.

TABLE 1. Environmental parameters for individual season mesocosm runs. Data are average (± SE) values for all mesocosms.

Season	Dates	Temperature (°C)	Salinity (‰)	pH
Spring 1997	May 13–June 6	19.1 (0.3)	9.6 (0.3)	8.2 (0.2)
Summer 1997	July 1–August 4	26.8 (0.2)	11.6 (0.4)	8.1 (0.1)
Late Summer 1997	August 19–September 22	24.7 (0.2)	13.8 (0.4)	8.1 (0.1)
Spring 1998	April 14–May 18	17.2 (0.3)	7.0 (0.0)	8.5 (0.1)

respond to increased loads of nutrients and trace elements is to make an integrative measure of its ecological properties. Net ecosystem metabolism is one of the few easily measured integrative properties (Odum and Hoskin 1958); it provides insight into the production and consumption of organic matter through photosynthesis and respiration, respectively. The balance between these two processes determines whether a system is a net producer or consumer of organic matter, affecting the amount of carbon that is stored and exported from a system (Odum 1971). In addition, the balance between photosynthesis and respiration plays a key role in determining whether a system is a net source or sink for carbon dioxide (Schindler et al. 1997; del Giorgio et al. 1999; Carpenter et al. 2001). The coupling between autotrophic and heterotrophic processes can also affect the amount and rate at which carbon is incorporated into lower trophic levels and transferred up the food web (Cole et al. 1988; Findlay et al. 1991; del Giorgio et al. 1999). This coupling can influence the structure, composition, and dynamics of food webs (del Giorgio et al. 1999). Nutrients and trace elements, alone and in combination, may change the balance and coupling between these processes.

The goal of this study was to examine the effects of nutrients and trace elements on whole system photosynthesis, respiration, and metabolism in a series of experimental estuarine food webs of increasing trophic complexity in twenty 1-m^3 mesocosms. The model for our simplified food web was the Patuxent River estuary, a Maryland tributary of the Chesapeake Bay. Nutrients (nitrogen and phosphorus) and trace elements (mix of arsenic, copper, and cadmium) were added alone and in combination during four mesocosm runs spanning spring 1997 to spring 1998. These pollutants were specifically chosen because they are currently of concern in the Patuxent River estuary and Chesapeake Bay (D'Elia et al. 1986; Sanders et al. 1987; Riedel et al. 1995, 1998, 2000). The research presented here is part of the COmplexity And STressors in Estuarine Systems (COASTES) project.

Materials and Methods

Experimental Design

The experimental design used in this study is that described by Breitburg et al. (1999) with minor modifications. The experiments were conducted in twenty 1-m^3 cylindrical fiberglass mesocosms (1.07-m diam × 1.22-m height) and carried out from spring 1997 to spring 1998 (Table 1). A factorial, randomized block design was used for the experiment, and the mesocosms were set up in a completely crossed design. There were 2 levels of nutrient additions × 2 levels of trace element additions × 5 levels of trophic level complexity. Using this experimental design, the entire data set could be statistically analyzed with the experimental run as the blocking factor. Main effects and 2-way interactions could also be analyzed for each of the four individual mesocosm runs.

The mesocosms were contained in outdoor raceways with flowing water from the Patuxent River estuary. This design maintained the mesocosm water temperature close to that of the estuary. Each mesocosm was filled with water from the Patuxent River estuary, screened through 35-μm mesh plankton nets. Mesocosms were maintained in a continuous flow-through mode in which 10% of the mesocosm water was exchanged daily for 1-μm filtered estuarine water. Mesocosm water was mixed by a PVC 4-blade paddlewheel. The paddlewheels rotated at 2 RPM and changed the direction of their rotation every 6 h. The interior surface of the mesocosm walls was lined with opaque PVC to prevent light penetration. Liners were removed weekly and cleaned to limit the effects of epiphytes on pelagic processes.

All mesocosms contained sandy sediments that were collected in bulk from the shallow sub-tidal Patuxent River estuary. The sediments were sieved and then heat-treated to kill macrofauna. Treated sediments were homogenized by stirring and then transferred to cylindrical (0.5-m diam and 0.15-m deep) PVC sediment trays. The sediment tray in the mesocosm yielded a sediment area:water column volume similar to that in a 4-m depth water column in the estuary.

The experiment was conducted over a 35-d period. During the initial 7 d, the mesocosms were allowed to acclimate. On day 7 and 8, nutrient and trace element loadings were elevated to target levels in the appropriate mesocosms (Table 2). After day 8, nutrient and trace element loading conditions were maintained by continuously pumping these contaminants into the mesocosms. Nutrient

TABLE 2. Nutrient loads and trace element concentrations in control and treatment mesocosms. Target nitrogen (N) and phosphorus (P) loading rates in the treatment mesocosms were 1.6 and 0.1 mmol d^{-1}, respectively. N and P were added in the forms of NO_3^- and PO_4^{3-}. Target concentrations for arsenic (As), copper (Cu), and cadmium (Cd) in the treatment mesocosms were 12.5, 7.5, and 0.5 μg l^{-1}, respectively. Loads for silica (Si) and dissolved organic carbon (DOC) are also shown. Data are average (± SE) values for control and treatment mesocosms during the individual season runs.

	Spring 1997		Summer 1997		Late Summer 1997		Spring 1998	
	Control	Treatment	Control	Treatment	Control	Treatment	Control	Treatment
N load (mmol N d^{-1})	2.0 (0.3)	3.6 (0.3)	1.5 (0.2)	3.1 (0.2)	1.1 (0.1)	2.7 (0.1)	1.0 (0.2)	2.6 (0.2)
P load (mmol P d^{-1})	0.02 (0.00)	0.12 (0.00)	0.05 (0.00)	0.15 (0.00)	0.07 (0.00)	0.17 (0.00)	0.01 (0.00)	0.11 (0.00)
As conc. (μg As l^{-1})	0.31 (0.01)	11.46 (0.17)	0.54 (0.02)	10.77 (0.21)	0.85 (0.03)	12.03 (0.30)	0.55 (0.01)	12.08 (0.21)
Cu conc. (μg Cu l^{-1})	1.06 (0.06)	5.48 (0.10)	0.71 (0.02)	5.13 (0.10)	0.69 (0.04)	4.15 (0.01)	0.83 (0.04)	4.57 (0.15)
Cd conc. (μg Cd l^{-1})	0.07 (0.00)	0.18 (0.01)	0.04 (0.00)	0.11 (0.00)	0.02 (0.00)	0.08 (0.00)	0.01 (0.00)	0.17 (0.01)
Si load (mmol Si d^{-1})	2.5 (0.2)	2.5 (0.2)	4.2 (0.8)	4.2 (0.8)	3.2 (0.4)	3.2 (0.4)	1.0 (0.4)	1.0 (0.4)
DOC load (mmol C d^{-1})	24.7 (1.2)	24.7 (1.2)	24.5 (0.8)	24.5 (0.8)	33.3 (5.7)	33.3 (5.7)	30.7 (0.7)	30.7 (0.7)

tanks (+N) received 1.61 mmol NO_3^--N d^{-1} and 0.1 mmol PO_4^{3-}-P d^{-1} (Table 2). The NO_3^- and PO_4^{3-} loads to the +N tanks were 1.8–2.6 and 2.4–11 times higher than the loads to the control tanks, which received water from the Patuxent River. The NO_3^- load to the +N mesocosms was 1.3–1.6 times higher than the average dissolved inorganic nitrogen (DIN) loadings measured to the nearby surface layer of the Patuxent River from 1984 to 1995 (Hagy 1996; Breitburg et al. 1999). These nutrient loadings were chosen to simulate effects of potential change in land use and population in the Patuxent River estuary watershed while maintaining a 16:1 nitrogen:phosphorus ratio in the nutrient addition. Target trace element concentrations for the trace element treatments (+T) were 12.5, 7.5, and 0.5 μg l^{-1} for arsenic, copper, and cadmium, respectively (Table 2). Trace element concentrations in +T tanks were 3–37 times higher than those in the controls (Table 2); they were 2–48 times higher than the ambient concentrations measured in the river along a salinity gradient during routine monitoring of the COASTES program from May 1995 to October 1997 (Riedel et al. 2000), but were within the range of concentrations measured within the Chesapeake Bay system. These trace element concentrations were chosen to cause changes in the structure and function of lower trophic levels but not to cause acute mortality to higher organisms (Sanders and Riedel 1998).

Five levels of system complexity were tested in this study, building from simple to successively more complex systems. Trophic complexity was defined in this study as the hierarchical organization of an ecological system's food web. The complexity treatments were +plankton (includes phytoplankton, bacteria, and microzooplankton from the estuarine water that passed through the 35-μm mesh net), +zooplankton (meso and larger microzooplankton added to the plankton assemblage), +fish (juvenile fish added to zooplankton assemblage), +bivalves (bivalves added to fish assemblage), and +benthos (anemones added to bivalve assemblage). Mesozooplankton additions were dominated by calenoid copepods (Table 3); *Acartia tonsa* was the most abundant species during the 4 runs (> 95%). These copepods feed on phytoplankton and microzooplankton (Heinle 1966). The copepod densities used in the experiment were chosen to simulate the average density found in the Patuxent River. The fish used were juvenile mummichogs (*Fundulus heteroclitus*; Table 3). Mum-

TABLE 3. Starting density and size of copepods (density only), fish, bivalves, and benthic invertebrates in each mesocosm run. Data are average values (± SE) for mesocosms with that trophic complexity treatment (refer back to Methods for trophic complexity treatment descriptions). Density is measured as the number of individuals in a mesocosm. *Mya* were not included in the summer and later summer runs. na = not applicable.

	Spring 1997	Summer 1997	Late Summer 1997	Spring 1998
Adult copepod density	3.8 (0.5)	3.8 (0.4)	4.4 (0.2)	5.1 (0.2)
Fish density	9 (0)	7 (0)	8 (0)	7 (0)
Wet fish weight (g)	0.04 (0.00)	0.06 (0.00)	0.1 (0.00)	0.06 (0.00)
Oyster density	50 (0)	50 (0)	50 (0)	50 (0)
Oyster area (cm^2)	1.96 (0.03)	2.17 (0.06)	2.45 (0.06)	2.45 (0.08)
Macoma density	82 (1)	108 (1)	65 (0)	35 (1)
Macoma area (cm^2)	0.94 (0.03)	0.99 (0.01)	1.30 (0.01)	1.26 (0.03)
Mya density	13 (1)	na	na	9 (0)
Mya area (cm^2)	1.21 (0.03)	na	na	1.41 (0.04)
Anemone density	21 (4)	39 (4)	21 (2)	39 (3)
Anemone area (cm^2)	0.63 (0.03)	0.25 (0.02)	0.21 (0.02)	0.82 (0.06)

michogs are one of the most abundant shallow-water fish in Atlantic coast estuaries and are important as both prey and predators in estuarine food webs (Kneib 1986). They feed on benthic invertebrates, benthic algae, and zooplankton. The fish densities used in the experiment were sufficient to allow for the fish growth rate in the tanks to be based on several fish but to avoid the complete consumption of fish prey in the mesocosms (Breitburg et al. 1999). The bivalves added were the eastern oyster (*Crassostrea virginica*) and two types of clam (*Macoma balthica* and *Mya arenaria*; *Mya* in spring 1997 and 1998 only; Table 3). These species are common in the Patuxent River and adjacent Chesapeake Bay and were chosen because they represent different habitat types and/or trophic groups. Oysters are a key epifaunal species in Chesapeake Bay found on hard substrates; they are suspension feeders primarily consuming phytoplankton, but are capable of consuming other suspended foods such as detritus and attached bacteria (Crosby et al. 1990; Langdon and Newell 1990; Baldwin and Newell 1991). *Macoma* are an infaunal species found in muddy and sandy environments; they are both suspension and deposit feeders. *Mya* are also found in sandy environments and are suspension feeders. The anemones used were *Diadumene leucolena* (Table 3). *Diadumene* are an epifaunal species found on hard substrates and are predators feeding primarily on zooplankton. The criteria that were used to determine the initial fish densities in the experiment were also used for the bivalves and anemones. Oysters were glued to PVC panels and suspended vertically and clams were added to the sediment tray in the +bivalves and +benthos. Anemones were attached to PVC plates suspended vertically in the +benthos mesocosms.

Measurements

Consecutive dawn-dusk-dawn measurements of dissolved oxygen were made weekly for each mesocosm to calculate rates for whole system gross primary production (WS-GPP), respiration (WS-RESP), and net ecosystem metabolism (NEM; Odum and Hoskin 1958; Oviatt et al. 1986a,b, 1993, 1995). Dissolved oxygen measurements were made with a YSI oxygen probe (Model 57) that was air-calibrated daily. Changes in the dissolved oxygen concentrations from $dawn_1$-dusk-$dawn_2$ were the result of biological processes (photosynthesis and respiration) and diffusive exchange (D) with the atmosphere; measurements for calculating the diffusive flux of oxygen across the air-water boundary are described in the following paragraphs. Rates of whole system net primary production (NPP) were calculated from the increase in the dissolved oxygen concentration from $dawn_1$ to dusk ($NPP = (O_{2dusk} - O_{2dawn1}/T) + D$, where O_2 was the dissolved oxygen concentration measured at dusk and $dawn_1$, T was the number of hours between these measurements, and D was the diffusive flux across the air-water boundary). WS-RESP rates were calculated from the decrease in the dissolved oxygen concentration from dusk to $dawn_2$ ($WS\text{-}RESP = (O_{2dusk} - O_{2dawn2}/T) + D$). Rates for WS-GPP were estimated from the sum of the rates for NPP and WS-RESP (WS-GPP = NPP + WS-RESP). NEM was calculated over a 24-h period as the difference between WS-GPP and WS-RESP ($NEM = WS\text{-}GPP_{day} - WS\text{-}RESP_{24H}$, where $WS\text{-}GPP_{day}$ was the amount of gross primary production that occurred during daylight hours and $WS\text{-}RESP_{24H}$ was the amount of respiration that occurred over 24 h). For this calculation, we assumed that the whole system respiration rate was constant over a diurnal cycle ($RESP_{day} = RESP_{night}$). This assumption is in agreement with other literature (Carignan et al. 2000).

Measured dissolved oxygen fluxes were corrected for air-water oxygen diffusion based on measured rates of SF_6 air-water diffusion (Wanninkhof et al. 1987; Upstill-Goddard et al. 1990; Wanninkhof 1992). SF_6 was added to five randomly chosen mesocosms and the change in its concentration was measured daily during the third week of each experiment. Tanks were inoculated with 7.5 ml of deionized water with a SF_6 concentration of 0.24 mmol SF_6 l^{-1}. Tanks equilibrated at least 12 h before sampling. Duplicate water samples were collected at 0.5-m depth from the mesocosms and were analyzed within a week of collection.

Dissolved SF_6 concentrations in the water were analyzed using a headspace equilibrium method. A 2-ml sample was extracted from the headspace and injected into a Shimadzu gas chromatograph equipped with an electron capture detector (Shimadzu GC-8A). SF_6 was separated from other atmospheric gases using a molecular sieve column 5A at a flow rate of 26 ml min^{-1} with N_2 as a carrier gas and a column and detector temperature of 25°C and 300°C, respectively (modified from Wanninkhof et al. 1987).

Measured SF_6 fluxes were used to calculate SF_6 piston velocities using the equation $k = F/(C_w - C_o)$, where k represents the piston velocity, F the flux of SF_6 from the tanks, C_w the concentration of SF_6 in the water, and C_o the concentration of SF_6 in the water at equilibrium with the atmosphere (Upstill-Goddard et al. 1990). The equation was simplified by assuming that Co equals zero because the concentration of SF_6 added to the mesocosm is orders of magnitude greater than the concentration in the water if it were in equilibrium with the atmosphere (Cole and Caraco 1998). SF_6 piston ve-

locities were converted to O_2 piston velocities using a Schmidt number relationship

$$k_{O2} = (Sc_{O2}/Sc_{SF6})^{(-1/2)} \times k_{SF6},$$

where k_{O2} is the oxygen piston velocity, Sc_{O2} is the Schmidt number for oxygen, Sc_{SF6} is the Schmidt number for SF_6, and k_{SF6} is the piston velocity for SF_6 (Wanninkhof et al. 1987; Wanninkhof 1992). Schmidt numbers were obtained from Wanninkhof (1992). The oxygen piston velocities from the five tanks were averaged and used to estimate diffusive fluxes from all tanks. Our oxygen piston velocities ranged from 5.9 to 10.8 cm h^{-1} and are comparable to those reported for other estuaries (Marino and Howarth 1993; Raymond and Cole 2001). In spring 1998, diffusive fluxes were not measured; instead, diffusive fluxes calculated for spring 1997 were applied (10.8 cm h^{-1}). Temperature, salinity, and stirring conditions for spring 1998 were similar to those in 1997.

Diffusive fluxes (D) of oxygen were calculated using the following equation:

$$D = k_{O2} \times (O_2 - O_{2sat}),$$

where O_2 is the measured dissolved oxygen concentration in the water and O_{2sat} is what the dissolved oxygen concentration would be in the water if it was in equilibrium with the atmosphere.

WS-GPP, WS-RESP, and NEM were converted to units of carbon assuming a photosynthetic and respiratory quotient of 1 (Valiela 1995; Falkowski and Raven 1997).

Statistical Analysis

Analysis of variance (ANOVA) was used to examine the direct and interactive effects of nutrient additions (N), trace element additions (T), and system complexity (C) on WS-GPP, WS-RESP, and NEM. WS-GPP, WS-RESP, and NEM were normalized using a square root transformation. The statistical model used to analyze the data set for all four mesocosm experiments (runs) was: variable = run + N + T + C + N × T + N × C + T × C + N × T × C. For analyses of the individual runs, the three-way interaction term and mesocosm run were dropped. Full models were simplified by successively dropping non-significant interaction terms ($p > 0.25$) until the overall F statistic for the main effects was no longer improved by the simplification. Main effects were always retained in the model. Post hoc tests (Tukey's Studentized Range and Least-Square Means [LSM]) were used to determine significant differences among trophic complexity treatments and trophic complexity/stressor interactions. Statistical analyses were conducted using SAS (version 8: GLM). Data in figures and text are presented as means (± SE) of stressor treatments (controls, nutrients only, trace elements only, nutrients plus trace elements) unless otherwise noted.

TABLE 4. P-values for ANOVA examining the main effects and 2-way interactions of nutrient additions, trace element additions, and trophic complexity on WS-GPP, WS-RESP, and NEM. N, T, and C represent nutrients, trace elements, and trophic complexity, respectively. Analyses of entire experiment and individual runs are presented. na = not applicable.

Main Effects/ 2-Way Interactions	WS-GPP	WS-RESP	NEM
Overall			
N	0.0001	0.0042	0.0001
T	0.0001	0.2382	0.0403
C	0.3147	0.6878	0.6105
N×T	0.0226	na	0.0158
N×C	0.0284	na	na
Spring 1997			
N	0.0001	0.0001	0.0071
T	0.0001	0.0001	0.0419
C	0.0371	0.0756	0.3481
N×T	0.0001	0.0582	0.0057
N×C	0.0027	na	na
Summer 1997			
N	0.0001	0.0001	0.0001
T	0.0002	0.0092	0.5754
C	0.9583	0.3450	0.4721
Late Summer 1997			
N	0.0001	0.0001	0.0003
T	0.0004	0.0624	0.2991
C	0.8065	0.0006	0.6800
N×T	na	0.0345	0.1968
Spring 1998			
N	0.0001	0.0185	0.0010
T	0.2268	0.0144	0.8409
C	0.0114	0.1095	0.0592
N×T	0.0924	na	0.0356
N×C	0.0277	na	na

Results

Whole System Gross Primary Production

Nutrient additions significantly affected WS-GPP over the entire experiment (Table 4). It was approximately 2.3 fold higher in +N mesocosms relative to the controls (Table 5 and Fig. 1a). WS-GPP was also significantly higher in treatments with nutrient additions as compared to controls during the individual runs (Table 4). It was 2.6 (average of two springs), 1.7, and 3.2 times higher in the +N mesocosms as compared to the controls in the spring, summer, and late summer, respectively (Table 5).

Trace element additions significantly affected WS-GPP over the entire experiment (Table 4); however, the direction of the response they elicited during individual runs varied (Table 5). During the spring runs, WS-GPP in +T mesocosms was similar to that of the controls (Table 5). However,

TABLE 5. WS-GPP, WS-RESP, and NEM (mmol C m^{-3} d^{-1}) in different stressor treatments for the entire experiment and individual runs. Data are averages (± SE). +N, +T, and +N+T represent tanks receiving nutrient additions only, trace element additions only, and combined nutrient and trace element additions, respectively.

Treatment	WS-GPP	WS-RESP	NEM
Overall			
Control	40.7 (2.8)	57.9 (2.8)	−17.2 (1.8)
+N	95.0 (3.8)	65.9 (5.6)	29.1 (7.6)
+T	36.1 (2.5)	61.0 (2.7)	−24.9 (1.4)
+N+T	71.3 (2.6)	69.6 (2.7)	1.6 (3.1)
Spring 1997			
Control	40.4 (2.3)	54.3 (4.2)	−13.9 (2.1)
+N	109.0 (10.9)	30.6 (4.3)	78.5 (13.7)
+T	44.4 (3.4)	67.9 (2.2)	−23.5 (2.5)
+N+T	64.1 (5.5)	56.2 (2.7)	8.0 (7.8)
Summer 1997			
Control	57.5 (4.8)	69.0 (3.4)	−11.5 (1.4)
+N	97.7 (4.5)	95.3 (4.7)	2.4 (4.6)
+T	41.2 (2.0)	61.1 (4.2)	−19.9 (2.6)
+N+T	69.4 (1.8)	80.9 (4.8)	−11.6 (4.5)
Late Summer 1997			
Control	27.5 (1.6)	43.6 (1.2)	−15.9 (2.3)
+N	84.1 (3.7)	67.1 (2.0)	17.0 (3.4)
+T	19.0 (0.6)	43.7 (2.1)	−24.6 (2.0)
+N+T	66.3 (3.4)	63.1 (1.1)	3.2 (3.9)
Spring 1998			
Control	37.5 (1.0)	65.0 (3.7)	−27.5 (3.8)
+N	89.2 (5.3)	70.8 (3.4)	18.4 (4.8)
+T	39.7 (1.8)	71.1 (1.8)	−31.4 (0.5)
+N+T	85.2 (3.2)	78.2 (1.9)	7.0 (5.1)

during the summer and late summer runs, trace element additions alone decreased WS-GPP (Table 5). The response of WS-GPP to trace elements in +N+T mesocosms contributed to the overall significant response of WS-GPP to trace elements (Fig. 1b,c). In the +N+T tanks, there was a significant interaction between nutrients and trace elements over the entire experiment where trace elements reduced the nutrient-induced WS-GPP response (Fig. 1c). While this pattern was consistent across most runs, it was only significant during spring 1997 (Table 4).

Trophic complexity did not significantly affect WS-GPP over the entire experiment (Table 4). Although there was a significant effect of trophic complexity on WS-GPP during spring 1997 and 1998, Tukey's Studentized Range and LSM post hoc analyses revealed that there were either no significant differences among the trophic treatments (spring 1997) or that trophic complexity did not consistently affect WS-GPP (spring 1998). In contrast, there was a significant interaction between nutrients and trophic complexity over the entire experiment affecting WS-GPP (Table 4). In +N tanks, WS-GPP was lower in the +bivalves and +benthos treatments compared to that in the +plankton and +fish treatments (Fig. 2; LSM post hoc test).

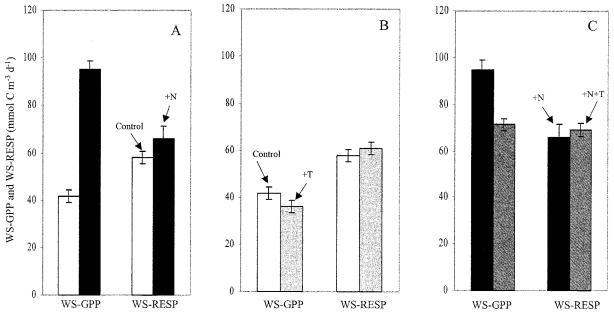

Fig. 1. Response of WS-GPP and WS-RESP to nutrient, trace element, and combined nutrient and trace element additions. A) Nutrients stimulated WS-GPP and to a lesser extent WS-RESP. B) Trace element additions alone decreased WS-GPP while not affecting WS-RESP. C) Trace element additions decreased WS-GPP in the presence of nutrients; they did not affect WS-RESP. Bars are overall averages (± SE). +N, +T, and +N+T represent tanks receiving nutrient additions only, trace element additions only, and combined nutrient and trace element additions, respectively.

Whole System Respiration

Nutrient additions significantly affected WS-RESP over the entire experiment (Table 4). It was 1.1 times higher in tanks with nutrients than in controls (Table 5 and Fig. 1a). In general, nutrient additions increased WS-RESP during the individual

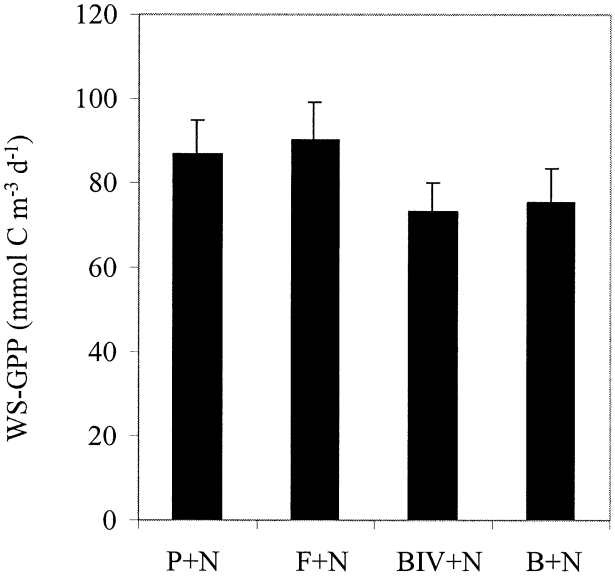

Fig. 2. Effects of trophic complexity on WS-GPP in nutrient tanks (+N). WS-GPP was lower in mesocosms with bivalves. Bars are overall averages (± SE). P, F, BIV, and B represent trophic complexity treatments +plankton, +fish, +bivalves, and +benthos, respectively.

runs. In the summer, late summer, and spring 1998, WS-RESP was 1.4, 1.5, and 1.1 times higher in tanks receiving nutrients relative to the controls (Table 5). In contrast, during spring 1997, nutrient additions decreased WS-RESP relative to the controls (Table 5).

Trace element additions did not significantly affect WS-RESP over the entire experiment (Table 4 and Fig. 1b). During the spring and summer runs trace elements did significantly affect WS-RESP (Table 4). In these runs, the direction of the response the trace elements elicited varied and the magnitude of the response was relatively small (Table 5). In the spring, WS-RESP was 1.2 (average of two springs) times higher in +T tanks compared to the controls (Table 5). In contrast, during summer 1997, WS-RESP in the +T tanks represented only 88% of the respiration observed in the controls (Table 5).

Trophic complexity did not significantly affect WS-RESP over the entire experiment except during late summer 1997 (Table 4). WS-RESP was significantly higher in the +bivalves (57.6 ± 5.1 mmol C m^{-3} d^{-1}; Tukey's Studentized Range post hoc test) treatment compared to the +plankton (55.2 ± 3.8 mmol C m^{-3} d^{-1}). It was also higher in the +benthos (59.2 ± 4.1 mmol C m^{-3} d^{-1}; Tukey's Studentized Range post hoc test) treatment compared to the +plankton (55.2 ± 3.8 mmol C m^{-3} d^{-1}), +zooplankton (52.9 ± 4.3 mmol C m^{-3} d^{-1}), and +fish (53.7 ± 4.4 mmol C m^{-3} d^{-1}).

Net Ecosystem Metabolism

Nutrient additions significantly affected NEM over the entire experiment and during the individual runs (Table 4). In general, +N mesocosms were autotrophic compared to the controls, which were heterotrophic (Table 5 and Fig. 3). Net autotrophy ranged from +2.4 to +78.5 mmol C m^{-3} d^{-1} in the +N mesocosms (Table 5). Net heterotrophy in the controls ranged from −11.5 to −27.5 mmol C m^{-3} d^{-1} (Table 5).

Trace elements significantly affected NEM over the entire experiment (Table 4). Their addition consistently resulted in greater net system heterotrophy compared to the controls (Fig. 3 and Table 5). In tests of individual mesocosm runs, however, trace elements only significantly affected NEM during spring 1997 (Table 4). It appears that the +N+T mesocosms contributed to the significant response of the NEM to trace elements (Table 4). In general, mesocosms receiving additions of trace elements and nutrients were less autotrophic than tanks receiving only nutrient additions (Table 5 and Fig. 3). Although the above pattern was consistent during all runs, the nutrient/trace element interaction was only significant during spring runs (Table 4).

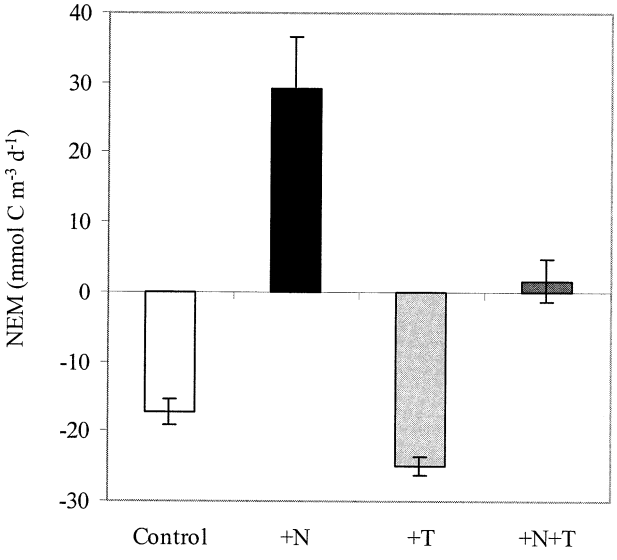

Fig. 3. Response of NEM to nutrient and trace element additions. Before stressor additions, the mesocosms were net heterotrophic. Nutrients alone made the tanks net autotrophic. Trace elements alone made the mesocosms slightly more heterotrophic than the controls. Combined additions of nutrients and trace elements resulted in WS-GPP = WS-RESP. Bars are overall averages (± SE). +N, +T, and +N+T represent tanks receiving nutrient additions only, trace element additions only, and combined additions of nutrients and trace elements, respectively.

Trophic complexity did not significantly affect NEM over the entire experiment or during the individual runs (Table 4).

Discussion

Estuarine Metabolic Fuel

Before the addition of either stressor, the COASTES mesocosms were net heterotrophic like the Patuxent River, suggesting that WS-RESP was in part fueled by allochthonous carbon inputs or by dissolved organic carbon (DOC) fluxes from the sediments (Hagy and Kemp 2002; Fig. 3 and Table 5). Calculations using measured benthic respiration rates from our control mesocosms (Laursen et al. 2002) and an equation relating sediment carbon oxidation (C_{ox}) to benthic DOC fluxes (BDF = $0.36(C_{ox})^{0.29}$; Burdige et al. 1999) suggest that less than 0.5% of the WS-RESP was fueled by BDF and that the majority was fueled by external carbon inputs. Allochthonous carbon entering the mesocosms was comprised of organic matter produced from both within the Patuxent River estuary and from the watershed. Most of this carbon was probably in the dissolved form because water entering the mesocosms passed through a 1-μm filter. The NEM represents 30% of the overall WS-RESP

in the control mesocosms; this is the minimum fraction of respiration that was fueled by allochthonous carbon if some of the autochthonous carbon produced within our tanks was buried or exported during the 24-h period over which our measurements were made. External inputs of carbon to rivers, lakes, and estuaries have been shown to support a similar percentage of WS-RESP in these environments (Smith and Hollibaugh 1993; Smith and Kemp 1995; Howarth et al. 1996; Cole et al. 2000). These studies do not distinguish how much of the respired allochthonous carbon is in the dissolved or particulate form. Our results suggest that DOC may be an important carbon source fueling estuarine metabolism. Estuarine DOC lability studies have found that ~ 30% of estuarine DOC is bioavailable (Servais et al. 1987; Aminot et al. 1990; Middelboe et al. 1992; Zweifel et al. 1993; reviewed in Wiegner and Seitzinger 2001). In the coastal Chesapeake Bay, DOC can comprise up to 80% of the total organic carbon (TOC; Fisher et al. 1998); our results suggest that 24% of the TOC may be removed by respiration of DOC.

Effects of Nutrients

Nutrient additions stimulated WS-GPP and to a lesser extent WS-RESP (Fig. 1a) resulting in net system autotrophy (Fig. 3). Previous studies have reported similar patterns in mesocosm and natural systems (Nixon et al. 1984; Oviatt et al. 1986a,b, 1995; D'Avanzo et al. 1996), yet few have examined why the magnitude of the response to nutrients by primary production and respiration differs. A dampened heterotrophic response to nutrient additions in the COASTES mesocosms may have occurred because of an inefficient or weakly coupled carbon flow between the autotrophs and heterotrophs. Uncoupling between these organisms can be related to the food web structure where there is top-down grazing of bacterioplankton (Carlson et al. 1996; Bird and Karl 1999) or a differential size partitioning of plankton community production and respiration (Smith and Kemp 2001). In addition, the quantity and quality of phytoplankton-derived carbon available to the bacteria may affect the coupling between WS-GPP and WS-RESP (Blight et al. 1995; Lochte et al. 1997; Carlson et al. 1998; Bird and Karl 1999; Serret et al. 1999). Bacterial utilization of allochthonous carbon may also contribute to an uncoupling between autotrophic and heterotrophic processes (Findlay et al. 1991; del Giorgio and Peters 1994; del Giorgio et al. 1999; Wikner and Hagströem 1999).

A dampened heterotrophic response to nutrients in the COASTES mesocosms may have resulted from several of the above factors. Nutrient additions weakly increased the rate of heterotrophic nanoflagellate (HNAN) grazing on bacteria during this (Gilmour and McManus unpublished data) and previous COASTES experiments (Breitburg et al. 1999). The heterotrophs may have also been consuming allochthonous carbon in addition to phytoplankton carbon. Approximately 30% of the carbon respired in our control tanks came from external sources; this percentage may represent the baseline respiration supported by allochthonous carbon in all our treatments. The size distribution of the plankton may also have played a role in the observed uncoupling. Previous studies in Chesapeake Bay have shown that the primary production driven by the > 3 μm fraction is not coupled to total plankton community respiration driven by the < 3 μm fraction (Smith and Kemp 2001). This relationship may explain why WS-GPP by large diatoms (Riedel et al. 2003) in our nutrient tanks responded to a greater extent to nutrients than WS-RESP (Fig. 1a).

Nutrient additions made the COASTES mesocosms net autotrophic (Fig. 3). Both the water column and benthos were net autotrophic in the +N tanks (calculated from our data and data from Laursen et al. 2002) suggesting that the net system production was not being respired, but rather exported and/or buried. Overall, we estimate that approximately 31% of the WS-GPP was exported from and/or buried in the +N tanks. Net system autotrophy has been observed in several temperate eutrophic estuaries (Nixon and Pilson 1984; Nixon et al. 1995; D'Avanzo et al. 1996; Kemp et al. 1997). Ten to 28% of the primary production in Narragansett Bay is estimated to be exported, while 5% to 10% is buried (Nixon and Pilson 1984; Nixon et al. 1995). In contrast, only 8% and 6% of the primary production in Chesapeake Bay is exported and buried, respectively (Kemp et al. 1997). NEM in coastal mesocosms has also been shown to switch from net heterotrophy to autotrophy with the addition of nutrients (Oviatt et al. 1986b). Ten to 15% of the net system production was exported from the mesocosms receiving nutrients, while 0% to 8% was buried (calculated from data in Oviatt et al. 1986b). Our results in conjunction with the literature suggest that estuaries receiving increased nutrient loads may be an important carbon source for downstream shelf areas (Odum 1971; Nixon and Pilson 1984; Kemp et al. 1997).

Effects of Trace Elements

The effects of trace elements at the whole system scale were not as apparent or consistent until nutrients were added to the COASTES mesocosms. The response of WS-GPP to nutrients was significantly dampened in the presence of trace elements whereas respiration was unaffected suggesting that

autotrophs and heterotrophs were affected differently by these stressors (Fig. 1c). Trace element effects on phytoplankton are well established (Sunda 1988–1989, 1994). They affect phytoplankton community composition, phytoplankton carbon, chlorophyll *a* concentration, primary production, and cell density (Sanders and Cibik 1985a; Brand et al. 1986; Sanders and Riedel 1998). In the +N COASTES mesocosms, the effect of trace elements were more pronounced; this may have occurred because some trace elements, like arsenate, are taken up by the same mechanisms as phosphate in phytoplankton (Planas and Healy 1978; Sanders 1979; Sanders and Windom 1980). The phytoplankton species most affected by trace elements were centric diatoms (Riedel et al. 2003); previous experiments have reported similar results (Sanders and Vermersch 1982; Sanders and Cibik 1985b; Sanders et al. 1989; Breitburg et al. 1999). However, some studies have found that certain diatoms are less sensitive to trace elements illustrating that effects of these contaminants are species-specific (Brand et al. 1986). In the context of the estuary, our results suggest that elevated trace element loads may dampen the response of primary production to nutrients, lessening the effects of eutrophication.

Heterotrophic bacteria are major contributors to estuarine respiration (Williams 1981; Hopkinson et al. 1989; Sand-Jensen et al. 1990; Sampou and Kemp 1994). In the COASTES mesocosms, effects of trace elements on these heterotrophs varied in +N+T tanks; bacterial cell density significantly decreased while bacteria production was unaffected (Gilmour and Ward unpublished data). Their experiments using ^{14}C-labelled substrates during the spring 1998 suggest that at some point in time trace elements decreased bacterial growth efficiency. WS-RESP and bacterial measurements together suggest that trace elements (with and without nutrients) may have decreased bacterial growth efficiency resulting in a higher respiration per cell. Although bacterial production did not change when trace elements were added, it is possible that proteins synthesized were being used to contend with high toxic trace element concentrations instead of being allocated for growth, resulting in a lower bacterial growth efficiency. Mechanisms used by bacteria to cope with these contaminants include internal and external elemental precipitation, syntheses of element binding/chelating organic compounds, and methylation (Gadd and Griffiths 1978).

Effects of Trophic Complexity

Food web structure has been shown to affect the primary productivity of some aquatic ecosystems (Elmgren et al. 1980; Carpenter et al. 1987; Alpine and Cloern 1992; Heath et al. 1995; Caraco et al. 1997; Carpenter et al. 2001). In our study, WS-GPP was significantly affected by trophic complexity in +N tanks (Table 4); it was lower in mesocosms with bivalves (both +bivalves and +benthos) relative to those without bivalves (Fig. 2). Measurements of ^{14}C-primary production and chlorophyll *a* were also significantly affected by trophic complexity, even though the effects were not exclusive to +N tanks (Breitburg unpublished data). Our results are not surprising given the fact that suspension-feeding bivalves have been shown to markedly decrease primary production in nutrient-rich aquatic environments (e.g., Cloern 1982; Cohen et al. 1984; Alpine and Cloern 1992; Caraco et al. 1997). In the Patuxent River estuary, it is estimated that suspension-feeding bivalves can remove up to 40% of the estuarine primary production (Gerritsen et al. 1994). In a marine mesocosm study, phytoplankton bloomed in mesocosms where the fuel oil had markedly reduced the number of the benthic suspension feeders in contrast to the controls (Elmgren et al. 1980). However, effects of suspension-feeding bivalves on primary production are not ubiquitous. In a study using *Mercenaria mercenaria,* clam additions increased ^{14}C-primary production relative to the controls due to an increased flux of dissolved inorganic nitrogen from the benthos (Doering et al. 1986).

Effects of trophic complexity on WS-RESP have not been as extensively studied as those for primary production. However, food web structure has been shown to affect pelagic and benthic respiration (Murphy and Kremer 1985; Doering et al. 1987; Pace and Cole 2000). In our study, trophic complexity did not significantly affect WS-RESP except during late summer 1997 when WS-RESP was significantly higher in treatments with bivalves (both +bivalves and +benthos). Benthic respiration was also significantly higher in treatments with bivalves during this run (Laursen et al. 2002). Increased WS-RESP and benthic respiration may be attributed to suspension-feeding bivalves, which accelerate carbon export from the water column to the benthos via filtration and defecation (Haven and Morales-Alamo 1966, 1972; Doering et al. 1986; Grant et al. 1995; Roditi et al. 1997). Fecal and pseudo fecal pellets may stimulate benthic microbial growth and, accordingly, respiration by maintaining a logarithmic growth phase of bacteria on the pellets (Dame 1996). Up to 20% of the increased oxygen consumption in the sediments relative to the controls in the COASTES mesocosms could be accounted for by respiration of the bivalves located on the sediments (Laursen et al. 2002). Similarly, in Colorado Lagoon, California,

M. mercenaria respiration was estimated to be 68% of the benthic respiration (Murphy and Kremer 1985).

Like WS-RESP, effects of trophic complexity on NEM have not been extensively examined. To our knowledge, there is only one other study that has examined trophic complexity effects on NEM. In that study, food web structure affected NEM in lakes receiving nutrient additions (Cole et al. 2000). The response of NEM to nutrients was top-down controlled; lakes were net autotrophic with a piscivorous food web and net heterotrophic with a planktivorous food web (Cole et al. 2000). Trophic complexity did not significantly affect NEM in our study although its effects on WS-GPP were detected in +N mesocosms (Table 4). Effects of trophic complexity on NEM in +N tanks may have not been detected because the magnitude of the NEM response to nutrients was much greater than the response of WS-GPP to trophic complexity.

Implications for Estuarine Systems

Human activities in watersheds and along the coast are resulting in increased export of nutrients and trace element contaminants to estuaries. These activities may also be changing the estuarine food web structure and dynamics. Our results suggest that these pollutants, alone and in combination, as well as the trophic complexity of the system, may affect the production, consumption, and flow of organic carbon in coastal areas. Nutrients enhanced WS-GPP and to a lesser extent WS-RESP in our system, shifting it from net heterotrophy to net autotrophy. The resulting net production was exported from or buried in the system. Bivalves also significantly reduced this nutrient-stimulated production. The addition of trace elements alone did not affect WS-GPP and WS-RESP to the same extent as nutrients and their effects were more variable, though additions of trace elements alone consistently made the system more net heterotrophic than the controls. When trace elements were added with nutrients, the nutrient-enriched system became less autotrophic. The effect of trace elements on NEM occurred primarily through reductions in WS-GPP rather than increases in WS-RESP. Our results suggest that the interactions between nutrient and trace element stressors on NEM are not additive. Both the direction and magnitude of the autotrophic and heterotrophic response to these stressors are different.

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Literature Cited

Alpine, A. E. and J. E. Cloern. 1992. Trophic interactions and direct physical effects control phytoplankton biomass and production in an estuary. *Limnology and Oceanography* 37:946–955.

Aminot, A., M. A. El-Sayed, and R. Kerouel. 1990. Fate of natural and anthropogenic dissolved organic carbon in the macrotidal Elorn estuary (France). *Marine Chemistry* 29:255–275.

Baldwin, B. S. and R. I. E. Newell. 1991. Omnivorous feeding by planktotrophic larvae of the eastern oyster *Crassostrea virginica*. *Marine Ecology Progress Series* 78:285–301.

Beers, J. R., G. L. Stewart, and K. D. Hoskins. 1977. Dynamics of micro-zooplankton populations treated with copper: Controlled ecosystem pollution experiment. *Bulletin of Marine Science* 27:66–79.

Bird, D. F. and D. M. Karl. 1999. Uncoupling of bacteria and phytoplankton during the austral spring bloom in Gerlache Strait, Antarctic Peninsula. *Aquatic Microbial Ecology* 19:13–27.

Blight, S. P., T. L. Bentley, D. Lefevre, C. Robinson, R. Rodrigues, J. Rowlands, and P. leB. Williams. 1995. Phasing of autotrophic and heterotrophic plankton metabolism in a temperate coastal ecosystem. *Marine Ecology Progress Series* 128: 61–75.

Brand, L. E., W. G. Sunda, and R. R. L. Guillard. 1986. Reduction of marine phytoplankton reproduction rates by copper and cadmium. *Journal of Experimental Marine Biology and Ecology* 96:225–250.

Breitburg, D. L., J. W. Baxter, C. A. Hatfield, R. W. Howarth, C. G. Jones, G. M. Lovett, and C. Wigand. 1998. Understanding effects of multiple stressors: Ideas and challenges, p. 416–431. *In* M. L. Pace and P. M. Groffman (eds.), Successes, Limitations, and Frontiers in Ecosystem Science. Springer-Verlag, New York.

Breitburg, D. L., J. G. Sanders, C. C. Gilmour, C. A. Hatfield, R. W. Osman, G. F. Riedel, S. P. Seitzinger, and K. G. Sellner. 1999. Variability in responses to nutrients and trace elements, and transmission of stressor effects through an estuarine food web. *Limnology and Oceanography* 44:837–863.

Burdige, D. J., W. M. Berelson, K. H. Coale, J. McManus, and K. S. Johnson. 1999. Fluxes of dissolved organic carbon from California continental margin sediments. *Geochimica et Cosmochimica Acta* 63:1507–1515.

Caraco, N. F., J. J. Cole, P. A. Raymond, D. L. Strayer, M. L. Pace, S. E. G. Findlay, and D. T. Fischer. 1997. Zebra mussel invasion in a large turbid river: Phytoplankton response to increased grazing. *Ecology* 78:588–602.

Carignan, R., D. Planas, and C. Vis. 2000. Planktonic production and respiration in oligotrophic Shield lakes. *Limnology and Oceanography* 45:189–199.

Carlson, C. A., H. W. Ducklow, D. A. Hansell, and W. O. Smith, Jr. 1998. Organic carbon partitioning during spring phytoplankton blooms in the Ross Sea polynya and the Sargasso Sea. *Limnology and Oceanography* 43:375–386.

Carlson, C. A., H. W. Ducklow, and T. D. Sleeter. 1996. Stocks and dynamics of bacterioplankton in the northwestern Sargasso Sea. *Deep-Sea Research II* 43:491–515.

Carpenter, S. R., J. J. Cole, J. R. Hodgson, J. F. Kitchell, M. L. Pace, D. Bade, K. L. Cottingham, T. E. Essington, J. N. Houser, and D. E. Schindler. 2001. Trophic cascades, nutrients, and lake productivity: Whole-lake experiments. *Ecological Monographs* 71:163–186.

Carpenter, S. R., J. F. Kitchell, J. R. Hodgson, P. A. Cochran, J. J. Elser, M. M. Elser, D. M. Lodge, D. Kretchmer, X. He, and C. N. von Ende. 1987. Regulation of lake primary productivity by food web structure. *Ecology* 68:1863–1875.

Cloern, J. E. 1982. Does the benthos control phytoplankton biomass in South San Francisco Bay? *Marine Ecology Progress Series* 9:191–202.

Cohen, R. R. H., P. V. Dresler, E. J. P. Phillips, and R. L. Cory. 1984. The effects of the Asiatic clam, *Corbicula fluminea,* on phytoplankton of the Potomac River, Maryland. *Limnology and Oceanography* 29:170–180.

Cole, J. J. and N. F. Caraco. 1998. Atmospheric exchange of carbon dioxide in a low-wind oligotrophic lake measured by the addition of SF_6. *Limnology and Oceanography* 43:647–656.

Cole, J. J., S. Findlay, and M. L. Pace. 1988. Bacterial production in fresh and saltwater ecosystems: A cross-system overview. *Marine Ecology Progress Series* 43:1–10.

Cole, J. J., M. L. Pace, S. R. Carpenter, and J. F. Kitchell. 2000. Persistence of net heterotrophy in lakes during nutrient addition and food web manipulations. *Limnology and Oceanography* 45:1718–1730.

Crosby, M. P., R. I. E. Newell, and C. J. Langdon. 1990. Bacterial mediation in the utilization of carbon and nitrogen from detrital complexes by *Crassostrea virginica. Limnology and Oceanography* 35:625–639.

Dame, R. F. 1996. Ecology of Marine Bivalves: An Ecosystem Approach. CRC Press, New York.

D'Avanzo, C., J. N. Kremer, and S. C. Wainright. 1996. Ecosystem production and respiration in response to eutrophication in shallow temperate estuaries. *Marine Ecology Progress Series* 141:263–274.

del Giorgio, P. A. and R. H. Peters. 1994. Patterns in planktonic P : R ratios in lakes: Influence of lake trophy and dissolved organic carbon. *Limnology and Oceanography* 39:772–787.

del Giorgio, P. A., J. J. Cole, N. F. Caraco, and R. H. Peters. 1999. Linking planktonic biomass and metabolism to net gas fluxes in northern temperate lakes. *Ecology* 80:1422–1431.

D'Elia, C. F., J. G. Sanders, and W. R. Boynton. 1986. Nutrient enrichment studies in a coastal-plain estuary: Phytoplankton growth in large-scale continuous cultures. *Canadian Journal of Fisheries and Aquatic Sciences* 43:397–406.

Doering, P. H., J. R. Kelly, C. A. Oviatt, and T. W. Sowers. 1987. Effect of the hard clam *Mercenaria mercenaria* on benthic fluxes of inorganic nutrients and gases. *Marine Biology* 94:377–383.

Doering, P. H., C. A. Oviatt, and J. R. Kelly. 1986. The effects of the filter-feeding clam *Mercenaria mercenaria* on carbon cycling in experimental marine mesocosms. *Journal of Marine Research* 44:839–861.

Elmgren, R., G. A. Vargo, J. F. Grassle, J. P. Grassle, D. R. Heinle, G. Langlois, and S. L. Vargo. 1980. Trophic interactions in experimental marine ecosystems perturbed by oil, p. 779–800. *In* J. P. Giesy (ed.), Microcosms in Ecological Research. Technical Information Center, U.S. Department of Energy, Springfield, Virginia.

Falkowski, P. G. and J. A. Raven. 1997. Aquatic Photosynthesis. Blackwell Science, Malden, Massachusetts.

Findlay, S., M. L. Pace, D. Lints, J. J. Cole, N. F. Caraco, and B. Peierls. 1991. Weak coupling of bacterial and algal production in a heterotrophic ecosystem: The Hudson River estuary. *Limnology and Oceanography* 36:268–278.

Fisher, T. R., J. D. Hagy, and E. Rochelle-Newall. 1998. Dissolved and particulate organic carbon in Chesapeake Bay. *Estuaries* 21:215–229.

Förstner, U. 1983a. Metal concentrations in river, lake, and ocean waters, p. 71–109. *In* U. Förstner and G. T. W. Wittmann (eds.), Metal Pollution in the Aquatic Environment. Springer-Verlag, New York.

Förstner, U. 1983b. Assessment of metal pollution in rivers and estuaries, p. 395–423. *In* I. Thornton (ed.), Applied Environmental Geochemistry. Academic Press, New York.

Gadd, G. M. and A. J. Griffiths. 1978. Microorganisms and heavy metal toxicity. *Microbial Ecology* 4:303–317.

Gamble, J. C., J. M. Davies, and J. H. Steele. 1977. Loch Ewe bag experiment. *Bulletin of Marine Science* 27:146–175.

Gerritsen, J., A. F. Holland, and D. E. Irvine. 1994. Suspension-feeding bivalves and the fate of primary production: An estuarine model applied to Chesapeake Bay. *Estuaries* 17:403–416.

Grant, J., A. Hatcher, D. B. Scott, P. Pocklington, C. T. Schafer, and G. V. Winters. 1995. A multidisciplinary approach to evaluating impacts of shellfish aquaculture on benthic communities. *Estuaries* 18:124–144.

Haven, D. S. and R. Morales-Alamo. 1966. Aspects of biodeposition by oysters and other invertebrate filter feeders. *Limnology and Oceanography* 11:487–498.

Haven, D. S. and R. Morales-Alamo. 1972. Biodeposition as a factor in sedimentation of fine suspended solids in estuaries. *Geological Society of America, Inc. Memoir* 133:121–130.

Hagy, J. D. 1996. Residence times and net ecosystem processes in Patuxent River estuary. M.S. Thesis, University of Maryland, College Park, Maryland.

Heath, R. T., G. L. Fahnenstiel, W. S. Gardner, J. F. Cavaletto, and S.-J. Hwang. 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.

Heinle, D. R. 1966. Production of a calenoid copepod, *Acartia tonsa,* in the Patuxent River estuary. *Chesapeake Science* 7:59–74.

Hopkinson, C. S., B. Sherr, and W. J. Wiebe. 1989. Size fractionated metabolism of coastal microbial plankton. *Marine Ecology Progress Series* 51:155–166.

Howarth, R. W. 1988. Nutrient limitation of net primary production in marine ecosystems. *Annual Review of Ecology* 19:89–110.

Howarth, R. W., R. Schneider, and D. Swaney. 1996. Metabolism and organic carbon fluxes in the tidal freshwater Hudson River. *Estuaries* 19:848–865.

Jickells, T. D. 1998. Nutrient biogeochemistry of the coastal zone. *Science* 281:217–222.

Kemp, W. M., E. M. Smith, M. Marvin-DiPasquale, and W. R. Boynton. 1997. Organic carbon balance and net ecosystem metabolism in Chesapeake Bay. *Marine Ecology Progress Series* 150:229–248.

Klerks, P. L. and J. S. Levinton. 1989. Effects of heavy metals in a polluted aquatic ecosystem, p. 41–67. *In* S. A. Levin, M. A. Harwell, J. R. Kelly, and K. D. Kimball (eds.), Ecotoxicology: Problems and Approaches. Springer-Verlag, New York.

Kneib, R. T. 1986. The role of *Fundulus heteroclitus* in salt marsh trophic dynamics. *American Zoologist* 26:259–269.

Langdon, C. J. and R. I. E. Newell. 1990. Utilization of detritus and bacteria as food sources by two bivalve suspension-feeders, the oyster *Crassostrea virginica* and the mussel *Geukensia demissa. Marine Ecology Progress Series* 58:299–310.

Laursen, A. E., S. P. Seitzinger, R. DeKorsey, J. G. Sanders, D. L. Breitburg, and R. W. Osman. 2002. Multiple stressors in an estuarine system: Effects of nutrients, trace elements, and trophic complexity on benthic photosynthesis and respiration. *Estuaries* 25:57–69.

Lochte, K., P. K. Bjørnsen, H. Giesenhagen, and A. Weber. 1997. Bacterial standing stock and production and their relation to phytoplankton in the Southern Ocean. *Deep-Sea Research II* 44:321–340.

Marino, R. and R. W. Howarth. 1993. Atmospheric oxygen exchange in the Hudson River: Dome measurements and comparison with other natural waters. *Estuaries* 16:433–445.

MIDDELBOE, M., B. NIELSEN, AND M. SØNDERGAARD. 1992. Bacterial utilization of dissolved organic carbon (DOC) in coastal waters-determination of growth yield. *Archiv für Hydrobiologie Ergebnisse der Limnologie* 37:51–61.

MURPHY, R. C. AND J. N. KREMER. 1985. Bivalve contribution to benthic metabolism in a California lagoon. *Estuaries* 8:330–341.

NIXON, S. W., S. L. GRANGER, AND B. L. NOWICKI. 1995. An assessment of the annual mass balance of carbon, nitrogen, and phosphorus in Narragansett Bay. *Biogeochemistry* 31:15–61.

NIXON, S. W. AND M. E. Q. PILSON. 1984. Estuarine total system metabolism and organic exchange calculated from nutrient ratios: An example from Narragansett Bay, p.261–290. *In* V. S. Kennedy (ed.), The Estuary as a Filter. Academic Press, Orlando, Florida.

NIXON, S. W., M. E. Q. PILSON, C. A. OVIATT, P. DONAGHAY, B. SULLIVAN, S. SEITZINGER, D. RUDNICK, AND J. FRITHSEN. 1984. Eutrophication of a coastal marine ecosystem—An experimental study using the MERL microcosms, p. 105–135. *In* M. J. R. Fasham (ed.), Flows of Energy and Materials in Marine Ecosystems. Plenum Press, New York.

NRIAGU, J. O. 1990. Global metal pollution poisoning the biosphere? *Environment* 32:7–33.

ODUM, E. P. 1971. Fundamentals of Ecology, 3rd edition. W. B. Saunders Company, Philadelphia, Pennsylvania.

ODUM, H. T. AND C. M. HOSKIN. 1958. Comparative studies on the metabolism of marine waters. *Publications of the Institute of Marine Science* 5:16–46.

OVIATT, C. A., D. T. RUDNICK, A. A. KELLER, P. A. SAMPOU, AND G. T. ALMQUIST. 1986a. A comparison of system (O_2 and CO_2) and C-14 measurements of metabolism in estuarine mesocosms. *Marine Ecology Progress Series* 28:57–67.

OVIATT, C. A., A. A. KELLER, P. A. SAMPOU, AND L. L. BEATTY. 1986b. Patterns of productivity during eutrophication: A mesocosm experiment. *Marine Ecology Progress Series* 28:69–80.

OVIATT, C. A., P. H. DOERING, B. L. NOWICKI, AND A. ZOPPINI. 1993. Net system production in coastal waters as a function of eutrophication, seasonality, and benthic macrofaunal abundance. *Estuaries* 16:247–254.

OVIATT, C., P. DOERING, B. NOWICKI, L. REED, J. COLE, AND J. FRITHSEN. 1995. An ecosystem level experiment on nutrient limitation in temperate coastal marine environments. *Marine Ecology Progress Series* 116:171–179.

PACE, M. L. AND J. J. COLE. 2000. Effects of whole-lake manipulations of nutrient loading and food web structure on planktonic respiration. *Canadian Journal of Fisheries and Aquatic Sciences* 57:487–496.

PLANAS, D. AND F. P. HEALEY. 1978. Effects of arsenate on growth and phosphorus metabolism of phytoplankton. *Journal of Phycology* 14:337–341.

RAYMOND, P. A. AND J. J. COLE. 2001. Gas exchange in rivers and estuaries: Choosing a gas transfer velocity. *Estuaries* 24:312–317.

REEVE, M. R., M. A. WALTER, K. DARCY, AND T. IKEDA. 1977. Evaluation of potential indicators of sub-lethal toxic stress on marine zooplankton (feeding, fecundity, respiration, and excretion): Controlled ecosystem pollution experiment. *Bulletin of Marine Science* 27:105–113.

RIEDEL, G. F., G. R. ABBE, AND J. G. SANDERS. 1995. Silver and copper accumulation in two estuarine bivalves, the eastern oyster (*Crassostrea virginica*) and the hooked mussel (*Ischadium recurvum*) in the Patuxent River estuary, Maryland. *Estuaries* 18:445–455.

RIEDEL, G. F., G. R. ABBE, AND J. G. SANDERS. 1998. Temporal and spatial variations of trace metal concentrations in oysters from the Patuxent River, Maryland. *Estuaries* 21:423–434.

RIEDEL, G. F., J. G. SANDERS, AND D. L. BREITBURG. 2003. Seasonal variability in response of estuarine phytoplankton communities to stress: Linkages between toxic trace elements and nutrient enrichment. *Estuaries* 26:323–339.

RIEDEL, G. F., S. A. WILLIAMS, G. S. RIEDEL, C. C. GILMOUR, AND J. G. SANDERS. 2000. Temporal and spatial patterns of trace elements in the Patuxent River: A whole watershed approach. *Estuaries* 23:521–535.

RODITI, H. A., D. L. STRAYER, AND S. E. G. FINDLAY. 1997. Characteristics of zebra mussel (*Dreissena polymorpha*) biodeposits in a tidal freshwater estuary. *Archiv fur Hydrobiologie* 140:207–219.

ROSENBERG, R. 1985. Eutrophication—The future marine coastal nuisance? *Marine Pollution Bulletin* 16:227–231.

SAMPOU, P. AND W. M. KEMP. 1994. Factors regulating plankton community respiration in Chesapeake Bay. *Marine Ecology Progress Series* 110:249–258.

SANDERS, J. G. 1979. Effects of arsenic speciation and phosphate concentration on arsenic inhibition of *Skeletonema costatum (Bacillariophyceae)*. *Journal of Phycology* 15:424–428.

SANDERS, J. G. AND H. L. WINDOM. 1980. The uptake and reduction of arsenic species by marine algae. *Estuarine and Coastal Marine Science* 10:555–567.

SANDERS, J. G., J. H. BATCHELDER, AND J. H. RYTHER. 1981. Dominance of a stressed marine phytoplankton assemblage by a copper-tolerant pennate diatom. *Botanica Marina* 24:39–41.

SANDERS, J. G. AND P. S. VERMERSCH. 1982. Response of marine phytoplankton to low levels of arsenate. *Journal of Plankton Research* 4:881–893.

SANDERS, J. G. AND S. J. CIBIK. 1985a. Adaptive behavior of euryhaline phytoplankton communities to arsenic stress. *Marine Ecology Progress Series* 22:199–205.

SANDERS, J. G. AND S. J. CIBIK. 1985b. Reduction of growth rate and resting spore formation in a marine diatom exposed to low levels of cadmium. *Marine Environmental Research* 16:165–180.

SANDERS, J. G., S. J. CIBIK, C. F. D'ELIA, AND W. R. BOYNTON. 1987. Nutrient enrichment studies in a coastal-plain estuary: Changes in phytoplankton species composition. *Canadian Journal of Fisheries and Aquatic Sciences* 44:83–90.

SANDERS, J. G. AND S. J. CIBIK. 1988. Response of Chesapeake Bay phytoplankton communities to low levels of toxic substances. *Marine Pollution Bulletin* 19:439–444.

SANDERS, J. G., R. W. OSMAN, AND G. F. RIEDEL. 1989. Pathways of arsenic uptake and incorporation in estuarine phytoplankton and filter-feeding invertebrates *Eurytmora affinis, Balanus improvisus,* and *Crassostrea virginica. Marine Biology* 103:319–325.

SANDERS, J. G. AND G. F. RIEDEL. 1998. Metal accumulation and impacts in phytoplankton, p. 59–76. *In* W. J. Langston and M. J. Bebianno (eds.), Metal Metabolism in Aquatic Environments. Chapman and Hall, London, U.K.

SAND-JENSEN, K., L. M. JENSEN, S. MARCHER, AND M. HANSEN. 1990. Pelagic metabolism in eutrophic coastal waters during a late summer period. *Marine Ecology Progress Series* 65:63–72.

SAÑUDO-WILHELMY, S. A. AND G. A. GILL. 1999. Impact of the Clean Water Act on the levels of toxic metals in urban estuaries: The Hudson River estuary revisited. *Environmental Science and Technology* 33:3477–3481.

SCHINDLER, D. E., S. R. CARPENTER, J. J. COLE, J. F. KITCHELL, AND M. L. PACE. 1997. Influence of food web structure on carbon exchange between lakes and the atmosphere. *Science* 277:248–251.

SERRET, P., E. FERNÁNDEZ, J. A. SOSTRES, AND R. ANADÓN. 1999. Seasonal compensation of microbial production and respiration in a temperate sea. *Marine Ecology Progress Series* 187:43–57.

SERVAIS, P., G. BILLEN, AND M. C. HASCOËT. 1987. Determination of the biodegradable fraction of dissolved organic matter in waters. *Water Research* 21:445–450.

SMITH, E. M. AND W. M. KEMP. 1995. Seasonal and regional var-

iations in plankton community production and respiration for Chesapeake Bay. *Marine Ecology Progress Series* 116:217–231.

SMITH, E. M. AND W. M. KEMP. 2001. Size structure and the production/respiration balance in a coastal plankton community. *Limnology and Oceanography* 46:473–485.

SMITH, S. V. AND J. T. HOLLIBAUGH. 1993. Coastal metabolism and the oceanic organic carbon balance. *Review of Geophysics* 31:75–89.

SUNDA, W. G. 1988–1989. Trace metal interactions with marine phytoplankton. *Biological Oceanography* 6:411–442.

SUNDA, W. G. 1994. Trace metal/phytoplankton interactions in the sea, p. 213–247. *In* G. Bidoglio and W. Stumm (eds.), Chemistry of Aquatic Systems: Local and Global Perspectives. Kluwer Academic Publishers, Netherlands.

UPSTILL-GODDARD, R. C., A. J. WATSON, P. S. LISS, AND M. I. LIDDICOAT. 1990. Gas transfer velocities in lakes measured with SF_6. *Tellus* 42b:364–377.

VALIELA, I. 1995. Marine Ecological Processes, 2nd edition. Springer-Verlag, New York.

WANNINKHOF, R. 1992. Relationship between wind speed and gas exchange over the ocean. *Journal of Geophysical Research* 97: 7373–7382.

WANNINKHOF, R., J. R. LEDWELL, AND W. S. BROECKER. 1987. Gas exchange on Mono Lake and Crowley Lake, California. *Journal of Geophysical Research* 92:14,567–14,580.

WIEGNER, T. N. AND S. P. SEITZINGER. 2001. Photochemical and microbial degradation of external dissolved organic matter inputs to rivers. *Aquatic Microbial Ecology* 24:27–40.

WIKNER, J. AND Å. HAGSTRÖEM. 1999. Bacterioplankton intra-annual variability: Importance of hydrography and competition. *Aquatic Microbial Ecology* 20:245–260.

WILLIAMS, P. J. LEB. 1981. Microbial contribution to overall marine plankton metabolism: Direct measurements of respiration. *Oceanologica Acta* 4:359–364.

ZWEIFEL, U. L., B. NORRMAN, AND Å. HAGSTRÖEM. 1993. Consumption of dissolved organic carbon by marine bacteria and demand for inorganic nutrients. *Marine Ecology Progress Series* 101:23–32.

SOURCES OF UNPUBLISHED MATERIALS

GILMOUR, C. C. Unpublished data. The Academy of Natural Sciences, Estuarine Research Center, 10545 Mackall Road, St. Leonard, Maryland 20685.

HAGY, J. D. AND W. M. KEMP. 2002. Patuxent River, Maryland. LOICZ Biogeochemical Budgets, Land-Ocean Interactions in the Coastal Zone, International Project Office, Netherlands Institute for Sea Research, Texel (http://www.nioz.nl/loicz/).

MCMANUS, G. B. Department of Marine Sciences, University of Connecticut, Groton, Connecticut 06340-6097.

WARD, M. Unpublished data. The Academy of Natural Sciences, Estuarine Research Center, 10545 Mackall Road, St. Leonard, Maryland 20685.

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