

# Nitrogen cycling in sediments with estuarine populations of *Potamogeton perfoliatus* and *Zostera marina*

Jane M. Caffrey\*, W. Michael Kemp

Horn Point Environmental Laboratories, University of Maryland, PO Box 775, Cambridge, Maryland 21613, USA

**ABSTRACT:** Rates of nitrogen transformations and concentrations of extractable  $\text{NH}_4^+$  and  $\text{NO}_3^-$  (plus  $\text{NO}_2^-$ ) were measured in estuarine sediments vegetated with the submersed macrophytes *Potamogeton perfoliatus* and *Zostera marina*, and in adjacent bare sediments, 3 or 4 times during the growing season. Nitrification and denitrification potentials were measured in substrate-amended sediment slurries at 5 depth intervals to provide a measure of bacterial activity. In general, rates were significantly higher in vegetated compared to bare sediments. It appears that both plant species affected nitrogen transformations through several similar mechanisms, while the microbial community, in turn, regulated nitrogen available for plant growth. In *P. perfoliatus* beds, ammonification and potential nitrification rates were correlated. Both exhibited summer maxima coinciding with peak plant biomass and productivity. Although vertically integrated (0–12 cm) ammonification rates were about twice as high in vegetated than in bare sediments,  $\text{NH}_4^+$  pools were significantly lower, probably due to high plant nitrogen demand. In contrast, denitrification rates were highest in spring when  $\text{NO}_3^-$  concentrations peaked, and were significantly correlated to nitrification rates in both spring and fall. Denitrification was only about 20% of total  $\text{NO}_3^-$  reduction, suggesting that  $\text{NH}_4^+$  production from  $\text{NO}_3^-$  may be important in conserving nitrogen within the grassbed. In sediments with *Z. marina*, rates of ammonification, and nitrification and denitrification potentials each exhibited a distinct seasonal cycle, indicating that rates were not as tightly coupled as in *P. perfoliatus* beds. High ammonification rates exceeded plant demand leading to  $\text{NH}_4^+$  accumulation. Potential nitrification rates were highest in vegetated sediments during fall. Denitrification rates, which were also greater in vegetated than in bare sediments, were highest in spring when  $\text{NO}_3^-$  concentrations were high. Potential denitrification rates comprised about 10% of total  $\text{NO}_3^-$  reduction, indicating that  $\text{NO}_3^-$  reduction to  $\text{NH}_4^+$  dominated. The microbial communities responsible for key nitrogen transformations in the sediments were enhanced by both *P. perfoliatus* and *Z. marina*: ammonification by inputs of organic nitrogen; nitrification by release of  $\text{O}_2$  by plant roots; and denitrification by production of  $\text{NO}_3^-$ .

## INTRODUCTION

Submersed vascular plant beds, which occur along salinity and latitudinal gradients (Stevenson 1988), are important sources of primary production in shallow aquatic ecosystems. These plants represent important physical structures, which provide habitat and nursery areas for many animals (Kikuchi 1980) and enhance the deposition of particulate material (Fonseca et al. 1982, Ward et al. 1984, Fonseca & Fisher 1986). Interconnected roots and rhizomes also increase sediment stability (Orth 1977).

Submersed plants can influence nitrogen cycling in sediments by several direct and indirect mechanisms. Ammonification, the microbial production of  $\text{NH}_4^+$  by deamination of organic matter, may be enhanced in vegetated sediments by: (1) decomposition of allochthonous particulate organic material trapped within the grassbed (Kemp et al. 1984); (2) microbial breakdown of dissolved organic nitrogen released from plant roots (Jørgensen et al. 1981, Boon 1986, Smith et al. 1988); and (3) decomposition of senescent plant material (Kenworthy & Thayer 1984). Relatively high rates of ammonification within macrophyte beds, compared to unvegetated sediments, can be sufficient to supply the nitrogen required for growth of the plants (Iizumi et al. 1982, Dennison et al. 1987).

\* Present address: Institute of Ecology and Genetics, University of Aarhus, Ny Munkegade, DK-8000 Aarhus C, Denmark

Plant nitrogen requirements for growth may also be met through nitrogen fixation. In tropical seagrass beds, sediment nitrogen fixation has been estimated to provide from 20 to 50 % of plant nitrogen demand (Capone et al. 1979, Capone & Taylor 1980). The contribution of sediment nitrogen fixation in temperate grassbeds tends to be lower than that in tropical beds, usually less than 30 % (Lipschultz et al. 1979, Capone 1983). Submersed plants may enhance nitrogen fixation in sediments, particularly in oligotrophic tropical environments, by release of labile dissolved organic compounds (Capone et al. 1979, O'Neill & Capone 1989).

Nitrification and denitrification are important processes controlling the loss of nitrogen in sediments. Nitrification, the oxidation of  $\text{NH}_4^+$  to  $\text{NO}_3^-$ , is controlled primarily by  $\text{O}_2$  and  $\text{NH}_4^+$  supply (Henriksen & Kemp 1988), while denitrification, the reduction of  $\text{NO}_3^-$  to  $\text{N}_2$ , is controlled by  $\text{NO}_3^-$  and organic carbon supply (Koike & Sørensen 1988). Often these 2 processes are closely coupled in time and space (Jenkins & Kemp 1984). Relatively high nitrification activities in vegetated sediments have been attributed to release of oxygen from eelgrass roots and rhizomes (Iizumi et al. 1980). Denitrification rates may be stimulated by plant metabolism either directly through excretion of dissolved organic carbon, or indirectly by increased  $\text{NO}_3^-$  production from nitrification. Relatively high denitrification activities have been measured in various seagrass (Capone & Taylor 1980, Iizumi et al. 1980, Kaspar 1983) and freshwater macrophyte beds (Christensen & Sørensen 1986).

In this paper, we compare microbial nitrogen transformation processes and porewater pools for 2 submersed plant species, and adjacent unvegetated sediments, occurring in Chesapeake Bay. The 2 plant species selected for this study were redhead grass *Potamogeton perfoliatus* and eelgrass *Zostera marina*, both of which were historically abundant in Chesapeake Bay, although populations have declined in recent decades (Kemp et al. 1983). *P. perfoliatus*, which occurs in oligohaline to mesohaline regions (salinity 0 to 15‰), exhibits seasonal peaks of production and biomass in mid summer (Twilley et al. 1985). *Z. marina*, which is found in euryhaline areas of Chesapeake Bay, follows a bimodal seasonal production cycle with maximum rates in late spring and early fall, and a precipitous late-summer decline in biomass associated with high temperatures (Wetzel & Penhale 1983). No previous studies have compared ammonification, nitrification and denitrification activities in vegetated versus adjacent unvegetated sediments for any submersed plant species. We hypothesized that the distinctly different growth strategies and physiological characteristics of these 2 plant species would result in different patterns of sediment nitrogen cycling processes.

## MATERIALS AND METHODS

**Study area and sampling procedures.** Sediment cores were collected from areas in the middle of a *Potamogeton perfoliatus* bed and from adjacent bare areas in an experimental estuarine pond (area 350 m<sup>2</sup>, mean depth 1 m, 38°36' N, 76°14' W) located near the Choptank River, a tributary of Chesapeake Bay, USA (Twilley et al. 1985). Sediments were collected with replicate 8.75 cm diameter clear acrylic cores for measurement of extractable  $\text{NH}_4^+$  and porewater  $\text{NO}_3^-$  plus  $\text{NO}_2^-$  concentrations, as well as rates of ammonification, nitrification, and denitrification. Half of the cores were collected from vegetated areas and the other half from unvegetated areas. Triplicate cores were used for each set of nitrogen transformation measurements, and for porewater nutrient concentrations. Measurements were made over 5 depth intervals: 0–1, 1–2, 2–4, 4–8, 8–12 cm. Sediments from each layer were completely mixed for all experiments. All sediments were incubated in filtered (GF/F) water from the Choptank River (salinity 10 to 12‰). Experiments were conducted in July and September 1986, and May 1987.

Cores with *Zostera marina* and bare sediments were collected from the lower Eastern Shore of Virginia in an area of Chesapeake Bay (MLW depth of 1 m) known locally as Vaucluse Shores (37°25' N, 75°39' W) and transported back to the laboratory in coolers filled with ambient water. Sampling and experimental protocols were the same for *Z. marina* as for *Potamogeton perfoliatus*, except as described below. Measurements were made at 0–1, 1–2, 2–6, 6–10, 10–14 cm depth intervals. Choptank River water was used for these experiments with salinity adjusted to ambient levels at the *Z. marina* site (20 to 25‰) by adding artificial sea salt (Instant Ocean). Experiments were conducted in June, August and November 1986, and April 1987.

**Plant biomass and production.** Aboveground biomass from each core was rinsed with fresh water, dried for 24 h at 60°C, and weighed. On 2 occasions for *Potamogeton perfoliatus* (July, August) and 3 occasions for *Zostera marina* (April, June, August), triplicate sediment cores were collected and roots and rhizomes were separated from each of the 5 depth intervals, dried at 60°C and weighed.

Productivities of *Potamogeton perfoliatus* (10 cm apical stems) and *Zostera marina* (intact leaf bundles) were measured as apparent daytime  $\text{O}_2$  production in duplicate 300 ml BOD bottles. Leaves were cleaned free of loosely attached epiphytic material by hand prior to each incubation.  $\text{O}_2$  concentrations were measured with polarographic electrodes at 30 min intervals for 3 h under natural light and in the dark. Light levels were adjusted, with neutral density screening, to ca

200  $\mu\text{Einsteins m}^{-2} \text{s}^{-1}$ , which is above saturation (Dennison & Alberte 1985, Goldsborough & Kemp 1988). Measurements were made monthly from April to September. Net diel productivity and C/N ratio of the plant (Twilley et al. 1986, W. C. Dennison pers. comm.) were used to calculate the nitrogen demand.

**Nitrogen rate measurements.** Potential nitrification rates were measured with sediment slurries in flasks (Henriksen 1980) by incubating ca 2 g of sediment, 50 ml of filtered estuarine water for 24 h under aerobic conditions at 25°C on a shaker table.  $\text{NH}_4\text{Cl}$  was added to bring  $\text{NH}_4^+$  concentrations to 1 mM. At 2 h and 24 h, sediments from replicate flasks were centrifuged and the water was filtered and analyzed for  $\text{NO}_3^-$ . On several occasions,  $\text{NO}_3^-$  measurements made at 14, 24 and 56 h indicated that rates were linear over a 56 h period.

We also used sediment slurries to measure potential rates of denitrification (Sørensen 1978, Kaspar 1983). Approximately 5 g of sediment, 40 ml of 100  $\mu\text{M}$   $\text{NO}_3^-$  and 5 ml of acetylene-saturated water were incubated for 4 h under anaerobic conditions (water and headspace were purged with nitrogen gas) at 25°C with shaking. The bottles were sampled after 0, 2, and 4 h for nitrous oxide ( $\text{N}_2\text{O}$ ) and  $\text{NO}_3^-$  in 1986. Rates of  $\text{N}_2\text{O}$  accumulation were linear over this time period. Four hours was not sufficient to measure loss of  $\text{NO}_3^-$ , so subsequent incubations (April and May) were run for 0, 12 and 24 h to measure  $\text{NO}_3^-$  disappearance. Gas samples (4 ml) were taken from the headspace in each bottle and analyzed for  $\text{N}_2\text{O}$ . A Hewlett Packard gas chromatograph with a  $^{63}\text{Ni}$  electron capture detector, with nitrogen as a carrier gas at a 15 ml  $\text{min}^{-1}$  flow rate (oven temperature 60°C, detector temperature 300°C, inlet temperature 120°C) and a 2 m column packed with Porapak Q, was used to measure  $\text{N}_2\text{O}$ .

Ammonification rates were measured using a technique similar to that described by Aller & Yingst (1980). Approximately 10 g of sediment in 30 ml of water were incubated under anaerobic conditions at 25°C for 4 d on a shaker table. The samples were centrifuged, and water was filtered and analyzed for  $\text{NH}_4^+$  at 0 and 4 d to estimate net  $\text{NH}_4^+$  production. Two time course experiments were conducted with sampling at 0, 2, 3 and 4 d and 0, 4 and 9 d.  $\text{NH}_4^+$  production was linear over the 2 sampling periods, except at the 6–10 and 10–14 cm depth intervals where  $\text{NH}_4^+$  production was close to zero.

For all experiments, roots and rhizomes were not included in the sediment slurries to avoid uptake or leakage of compounds such as  $\text{NH}_4^+$  or DOC to or from plant tissues. Consequently, the bacterial community attached or immediately adjacent to roots and rhizomes was not included. However, on 4 occasions, sediments were scraped from roots and rhizomes and used to

measure potential nitrification, potential denitrification and ammonification rates.

**Porewater measurements.** Sediment samples (about 15 g wet sediment) were extracted with 20 ml of 1N KCl to determine extractable  $\text{NH}_4^+$  concentrations. These measurements include  $\text{NH}_4^+$  dissolved in the porewater and sorbed to sediment particles.  $\text{NO}_3^-$  plus  $\text{NO}_2^-$  (hereafter referred to as  $\text{NO}_3^-$ ) concentrations were also measured in the KCl extracts.  $\text{NH}_4^+$  and  $\text{NO}_3^-$  concentrations in KCl extracts and KCl blanks were analyzed with a Technicon II system.  $\text{NH}_4^+$  was determined by the phenol hypochlorite method (Solorzano 1969), and the automated cadmium reduction method was used for analysis of  $\text{NO}_3^-$  plus  $\text{NO}_2^-$  (APHA 1985). Concentrations of  $\text{NH}_4^+$  and  $\text{NO}_3^-$  are reported in terms of volume of porewater, while rates of nitrogen transformations are expressed in  $\text{nmol N cm}^{-3}$  (dry sediment)  $\text{h}^{-1}$ . Extractable  $\text{NH}_4^+$  concentrations are reported in terms of porewater volume because extractable  $\text{NH}_4^+$  is in equilibrium between sediment particles and porewater (Rosenfeld 1979) and would thus be readily available to the plants. Percent water was measured at all depth intervals.

We tested for differences in rates of nitrogen transformations between the vegetated and bare sediments each month using the Kruskal-Wallis analysis of variance for each nitrogen transformation (SYSTAT 1986). Probabilities (p) of 0.1 or less were considered significant and exact p values are usually reported. Non-parametric analysis of variance was used instead of parametric tests because of unequal variances, even after transformation of the variables.

## RESULTS

Rates of nitrogen transformations and concentrations of extractable  $\text{NH}_4^+$  and  $\text{NO}_3^-$  differed between vegetated sediments and adjacent bare sediments for both plant species. Results are presented first for *Potamogeton perfoliatus* and then for *Zostera marina*.

### *Potamogeton perfoliatus*

Ammonification rates were significantly greater in vegetated sediments compared to bare sediments ( $p < 0.05$ ), except in May. In bare and vegetated sediments, ammonification was usually highest at the surface (0–1 cm), and declined to 0 below 2 cm in bare sediments (Fig. 1). Rates within the *Potamogeton perfoliatus* beds were greatest in July at the 0–1 cm layer (71.4  $\text{nmol N cm}^{-3} \text{h}^{-1}$ ), with a second peak in the 4–8 cm layer (9.8  $\text{nmol N cm}^{-3} \text{h}^{-1}$ ). Root and rhizome biomass was concentrated in the top 8 cm (Fig. 2).

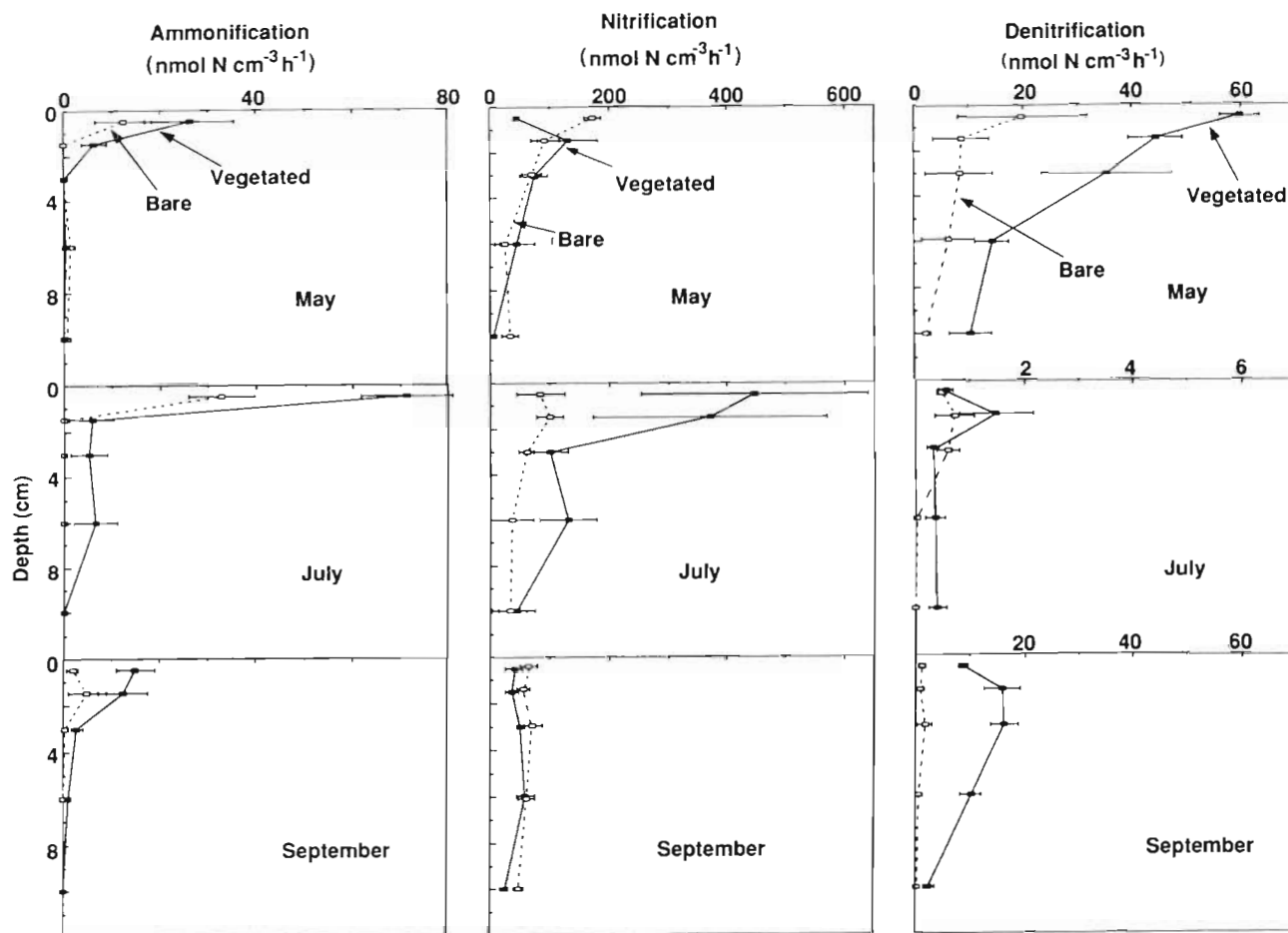


Fig. 1. Vertical profiles of rates of ammonification, potential nitrification and potential denitrification ( $\text{nmol cm}^{-3} \text{h}^{-1}$ ) within *Potamogeton perfoliatus* beds (closed boxes, continuous lines) and adjacent bare sediments (open boxes, dashed lines) in May, July and September. Means  $\pm$  standard errors ( $n = 3$ ) are given. Note that the scale for denitrification rates in July is 10 % of that in other months

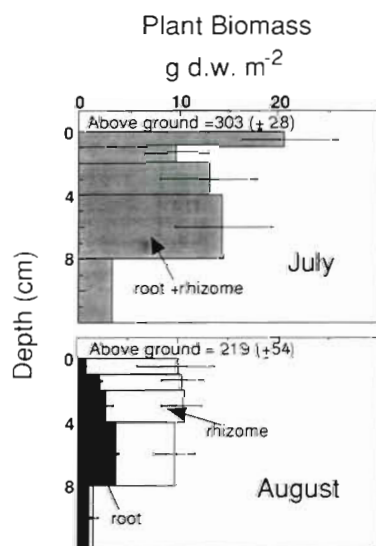


Fig. 2. *Potamogeton perfoliatus*. Root + rhizome biomass ( $\text{g d.w. m}^{-2}$ ) in (a) July and (b) August. Mean  $\pm$  standard errors ( $n = 3$ ) and biomass of aboveground plant parts are given

Ammonification rates, integrated over the top 12 cm, in vegetated sediments were 2 to 3 times those in bare sediments during summer and fall sampling periods, but only slightly higher in the spring (Table 1). Rates in bare and vegetated sediments were significantly different in July ( $p < 0.05$ ) and September ( $p < 0.003$ ), but not in May ( $p < 0.14$ ). Despite these higher ammonification rates, extractable  $\text{NH}_4^+$  concentrations were lower in the *P. perfoliatus* beds than in bare sediments in spring and summer sampling periods (Fig. 3), where  $\text{NH}_4^+$  concentrations increased with depth. Ammonification was significantly related to  $\text{NH}_4^+$  concentrations in the top centimeter ( $Y = 145 + 5.11X$ ,  $r = 0.69$ ).

Differences in potential nitrification between vegetated and bare sediments were most pronounced in July (Fig. 1) when potential nitrification had a similar temporal pattern as ammonification. During the summer sampling period, highest rates of nitrification activity were in the top 1 cm layer, reaching a maximum value of  $452 \text{ nmol N cm}^{-3} \text{h}^{-1}$  in vegetated sediments.

Table 1. Integrated depth profiles (0–12 cm) of potential nitrogen transformations (ammonification, nitrification, and denitrification) measured in sediment slurries from sediments vegetated with *Potamogeton perfoliatus* and adjacent bare sediments (mean ( $\pm$  SE) for  $n = 3$ )

	Ammonification	Nitrification	Denitrification
	( $\mu\text{mol m}^{-2} \text{h}^{-1}$ )		
Vegetated			
May	111 (38)	3223 (604)	638 (110)
Jul	264 (45)	4760 (1036)	10 (1)
Sep	148 (42)	3859 (752)	262 (15)
Bare			
May	102 (45)	3450 (851)	220 (111)
Jul	105 (22)	1727 (326)	6 (2)
Sep	40 (22)	2520 (336)	26 (10)

Vertical profiles revealed that nitrification potentials in vegetated sediments were 3 times those in bare sediments at the 4–8 cm layer (Fig. 1). Although potential nitrification rates in July were substantial below the

surface, there was no accumulation of  $\text{NO}_3^-$  at depth (2–12 cm) in vegetated sediments. However, surficial (0–1 cm)  $\text{NO}_3^-$  concentrations were always greater than overlying water concentrations.  $\text{NO}_3^-$  concentrations were different in vegetated and bare sediments (Fig. 3) in May and July. In sediments occupied by *Potamogeton perfoliatus*, highest nitrification rates occurred during sampling periods in summer, when macrophyte production was also high (Fig. 1, Table 1). These potential nitrification rates were significantly different between bare and vegetated sediments in July ( $p < 0.06$ ) and September ( $p < 0.003$ ), but not in May ( $p < 0.22$ ). Nitrification rates in vegetated sediments were significantly ( $p < 0.05$ ) correlated with porewater  $\text{NH}_4^+$  concentrations in the top 8 cm ( $Y = -49.0 + 1.3X$ ,  $r = 0.64$ ) and with ammonification rates at 0–12 cm ( $Y = 59.2 + 4.7X$ ,  $r = 0.67$ ). Similar correlations were not significant for the bare sediments.

The influence of *Potamogeton perfoliatus* on potential denitrification was different from its effect on potential nitrification or on ammonification rates.

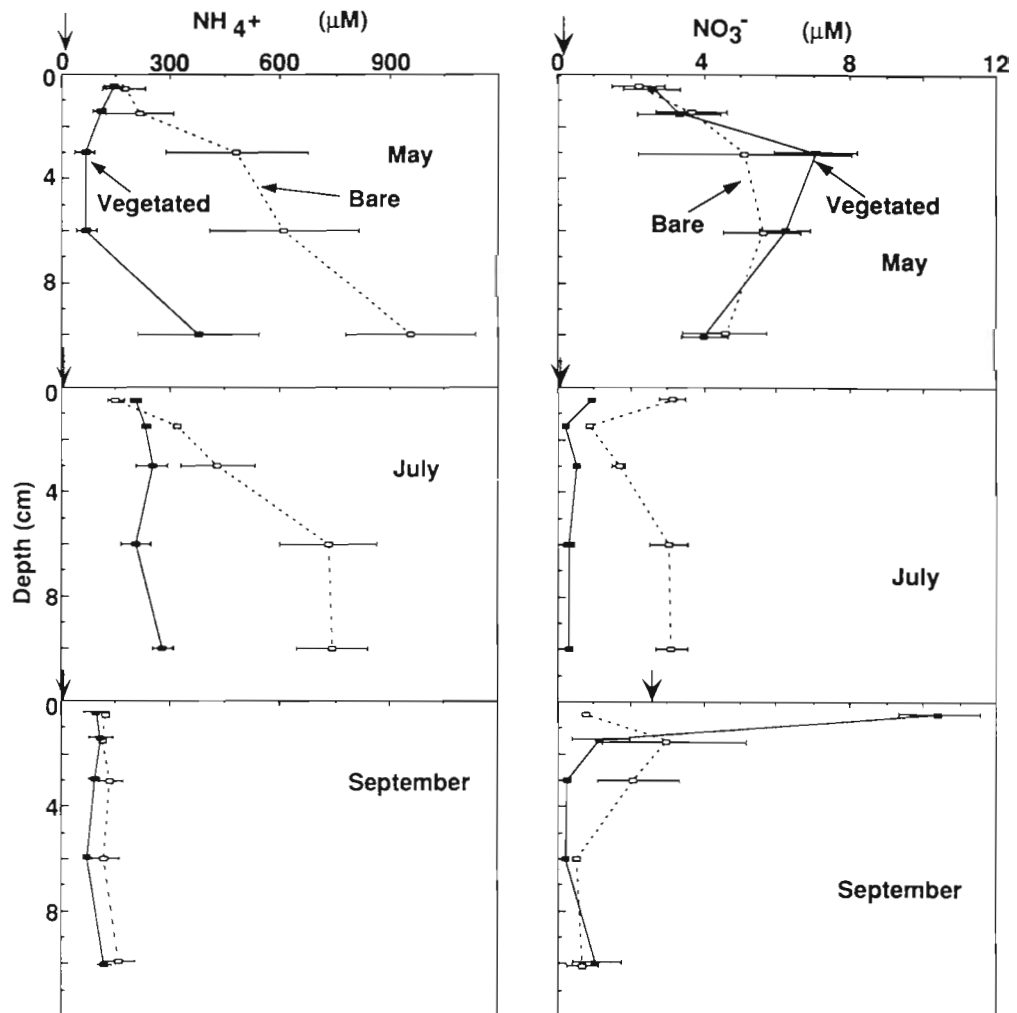


Fig. 3. Vertical profiles of extractable  $\text{NH}_4^+$  and  $\text{NO}_3^-$  plus  $\text{NO}_2^-$  concentrations ( $\mu\text{mol l}^{-1}$  pore water) within *Potamogeton perfoliatus* beds (closed boxes, continuous lines) and adjacent bare sediments (open boxes, dashed lines) in May, July and September. Means  $\pm$  standard errors ( $n = 3$ ) are given. Vertical arrows indicate overlying water concentrations



Denitrification potentials were generally lower than ammonification and nitrification rates, ranging from 0 to 60  $\text{nmol N cm}^{-3} \text{ h}^{-1}$ . Denitrification potentials had a subsurface maximum in July and September, while rates were highest at the surface and declined with depth in May. Rates were generally higher in vegetated than in unvegetated sediments and were significantly different ( $p < 0.001$ ) in May and September, but not in July ( $p < 0.21$ ). In contrast to the seasonal pattern of ammonification in *P. perfoliatus* beds, denitrification rates were greater in spring and fall and lower in summer (Fig. 1, Table 1). In vegetated sediments below 2 cm, potential denitrification and nitrification rates were correlated ( $p < 0.11$ ) in spring and fall ( $Y = 20.1 + 1.57X$ ,  $r = 0.71$ ).

In spring, total  $\text{NO}_3^-$  reduction, in sediment slurries, was higher in vegetated sediments than in bare sediments. Rates within the vegetated sediments were 260  $\text{nmol N cm}^{-3} \text{ h}^{-1}$  at the surface and 140  $\text{nmol N cm}^{-3} \text{ h}^{-1}$  at 6–10 cm, while rates within the adjacent, bare sediments were 45  $\text{nmol N cm}^{-3} \text{ h}^{-1}$  and showed little variability below the 0–1 cm layer (Fig. 4). The relation-

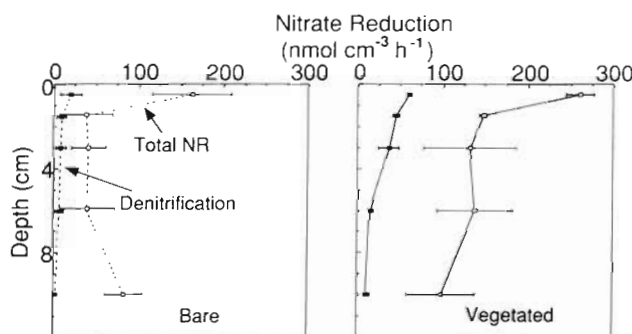


Fig. 4. Total nitrate reduction (open boxes) and denitrification (closed boxes) ( $\text{nmol cm}^{-3} \text{ h}^{-1}$ ) in *Potamogeton perfoliatus* beds (continuous lines) and adjacent bare sediments (dotted lines) in May

ship between total  $\text{NO}_3^-$  reduction and denitrification suggests that denitrification comprised about 20 % of total  $\text{NO}_3^-$  reduction ( $Y = -5.5 + 0.23X$ ,  $r = 0.85$ ).

### *Zostera marina*

Nitrogen transformations (ammonification, potential nitrification and potential denitrification) and concentrations ( $\text{NH}_4^+$  and  $\text{NO}_3^-$ ) were significantly ( $p < 0.10$ ) enhanced in *Zostera marina* sediments compared with adjacent, bare sediments (Figs. 5 and 6, Table 2). Ammonification rates were enhanced in sediments within the eelgrass beds during all sampling periods. The greatest differences in subsurface rates between bare and vegetated sediments occurred in summer,

with less of a difference in the fall and spring (Fig. 5). In June and August, rates were highest at the surface with a second peak in the 2–6 cm layer (Fig. 5); these subsurface peaks coincided with the zone of maximum root and rhizome biomass (Fig. 7). Rates in bare sediments decreased with depth. Ammonification rates were significantly different ( $p < 0.10$ ) between bare and vegetated sediments, except in April. Extractable  $\text{NH}_4^+$  concentrations during summer sampling periods were much greater in vegetated than bare sediments (Fig. 6).  $\text{NH}_4^+$  concentrations (X) and ammonification rates (Y) were positively correlated ( $p < 0.008$ ) for both vegetated ( $Y = 161.6 + 3.54X$ ,  $r = 0.59$ ) and bare sediments ( $Y = 64.4 + 11.9X$ ,  $r = 0.63$ ).

Potential nitrification rates were also greater in vegetated than bare sediments, except in November. These rates increased throughout the year and were at a maximum in November (Fig. 5, Table 2) despite relatively low  $\text{NH}_4^+$  concentrations at that time. Potential nitrification was significantly different between vegetated and bare sediments in June ( $p < 0.008$ ) and November ( $p < 0.03$ ), but not in April ( $p < 0.41$ ) or August ( $p < 0.12$ ).  $\text{NO}_3^-$  concentrations in sediments were relatively high in June and November at both bare and vegetated sites, in April at the vegetated site, and much greater than overlying water concentrations during all sampling periods (Fig. 6). Potential nitrification rates (X) and  $\text{NO}_3^-$  concentrations (Y) were significantly ( $p < 0.08$ ) correlated in vegetated sediments below the top centimeter ( $Y = 0.70 + 0.16X$ ,  $r = 0.47$ ).  $\text{NO}_3^-$  concentrations generally decreased with depth, while vertical profiles of nitrification potential exhibited subsurface maxima (1–2 cm) in April and November (Fig. 5). Nitrification potentials did not seem to be related to either  $\text{NH}_4^+$  concentrations or ammonification rates.

The enhancement of potential denitrification rates in vegetated versus bare sediments was similar to that observed for ammonification. Maximum denitrification rates were in the 0–1 cm layer except in August (Fig. 5). Rates were higher in spring and fall than in summer (Fig. 7, Table 2). Differences between bare and vegetated sediments were significant in all 4 months ( $p < 0.06$ ). Potential denitrification rates were positively correlated ( $p < 0.0001$ ) with  $\text{NO}_3^-$  concentrations in vegetated sediments ( $Y = 0.05 + 0.52X$ ,  $r = 0.94$ ), but not in bare sediments. Although  $\text{NO}_3^-$  concentrations and denitrification potentials were related, nitrification potentials and denitrification potentials were not. The vertical pattern for total  $\text{NO}_3^-$  reduction in vegetated sediments was very similar to that of denitrification in April, having high rates at the surface (240  $\text{nmol N cm}^{-3} \text{ h}^{-1}$ ) and a second peak at 6–10 cm (Fig. 8). Denitrification and  $\text{NO}_3^-$  reduction rates were significantly correlated ( $p < 0.05$ ) in vegetated ( $Y = -3.17 +$

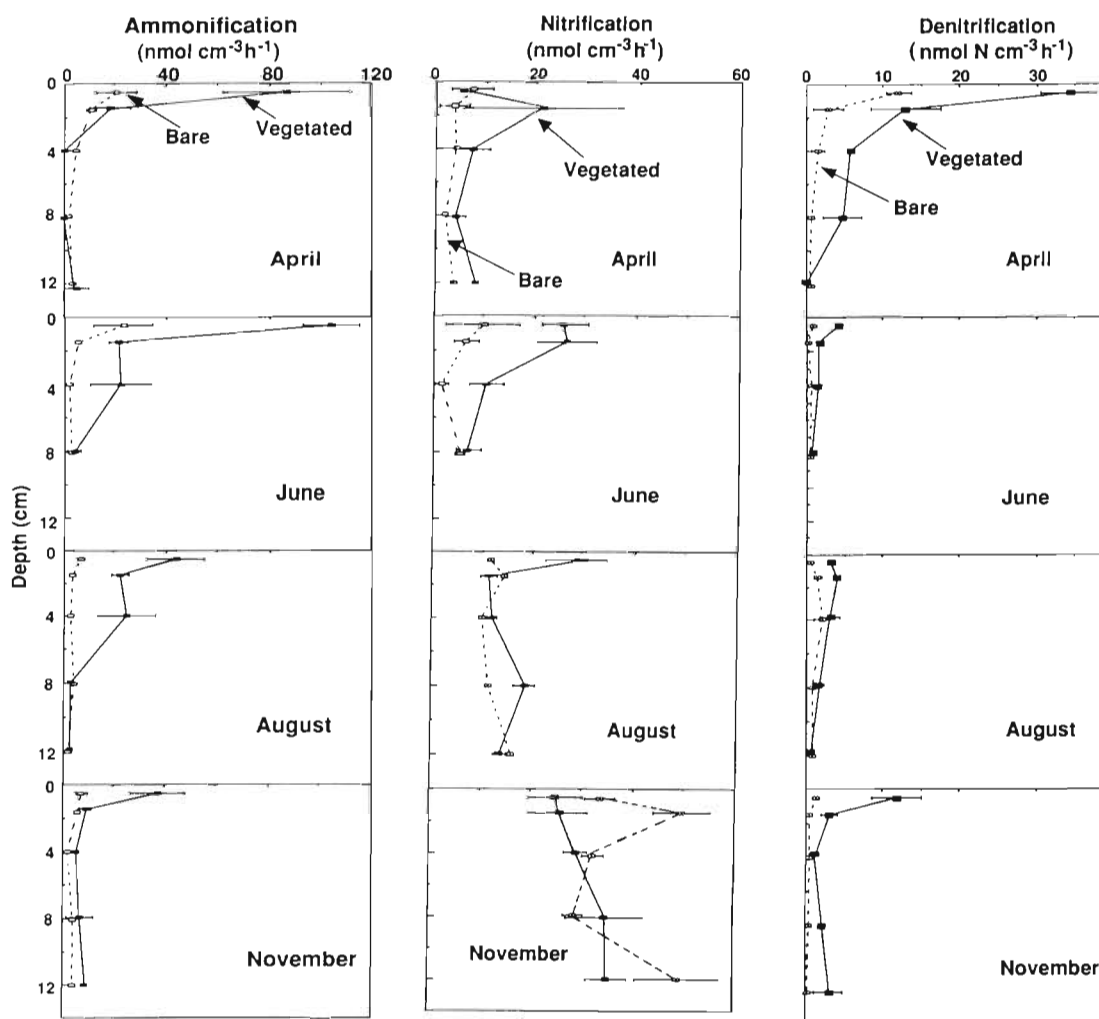


Fig. 5. Vertical profiles of ammonification, potential nitrification and potential denitrification rates (nmol cm<sup>-3</sup> h<sup>-1</sup>) within *Zostera marina* beds (closed boxes, continuous lines) and adjacent bare sediments (open boxes, dashed lines) in April, June, August, and November. Means  $\pm$  standard errors (n = 3) are given

0.13X,  $r = 0.88$ ), but not in bare sediments. In general, denitrification represented about 10% of total  $\text{NO}_3^-$  reduction in vegetated sediments, and a much smaller fraction of total  $\text{NO}_3^-$  reduction in bare sediments.

## DISCUSSION

### Nitrogen transformation rates

Results indicate that both *Potamogeton perfoliatus* and *Zostera marina* significantly influence nitrogen transformation processes and nitrogen pools in sediments. The seasonality of ammonification and nitrification potentials tended to follow the growth cycle of *P. perfoliatus*, while denitrification did not. Although there were strong seasonal patterns for the rates of nitrogen transformations in *Z. marina* beds, each pro-

cess exhibited a different seasonality. The observed rates were generally similar to those found in other systems containing submersed plants (Koike & Hattori 1978b, Iizumi et al. 1980, Kaspar 1983, Kepkay 1985, Boon et al. 1986a, b, Christensen & Sørensen 1986, Dennison et al. 1987, Short 1987; Table 3). The sediment nitrogen cycling rates for these 2 Chesapeake Bay plant populations were at the upper end of the range of values reported, particularly for nitrification and nitrate reduction.

Comparisons of these rates in vegetated and adjacent bare sediments may, in fact, underestimate the effect of the plant, since the bare sites were close to the grassbed and subject to the plants' influence. For the results presented, the bacterial community attached to rhizoplane was excluded, which may underestimate the macrophyte effect. On several occasions experiments were run using proximal sediments attached to

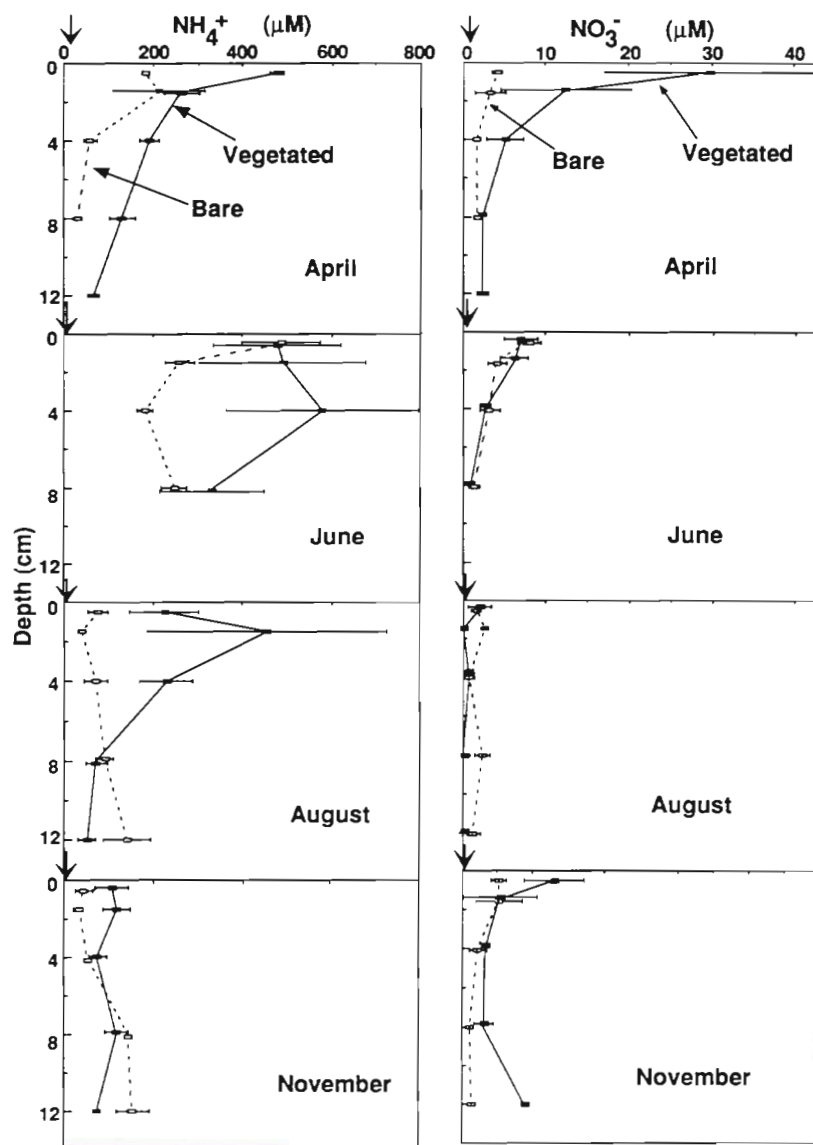


Fig. 6. Vertical profiles of extractable  $\text{NH}_4^+$  and  $\text{NO}_3^-$  plus  $\text{NO}_2^-$  concentrations ( $\mu\text{mol l}^{-1}$  pore-water) within *Zostera marina* beds (closed boxes, continuous lines) and adjacent bare sediments (open boxes, dashed lines) in April, June, August, and November. Means  $\pm$  standard errors ( $n = 3$ ) are given. Vertical arrows indicate overlying water concentrations

Table 2. Integrated depth profiles (0–14 cm) of potential nitrogen transformations (ammonification, nitrification, and denitrification) in sediments vegetated with *Zostera marina* and adjacent bare sediments (mean  $\pm$  SE) for  $n = 3$

	Ammonification	Nitrification	Denitrification
	( $\mu\text{mol l m}^{-2} \text{ h}^{-1}$ )		
Vegetated			
Apr	1094 (108)	410 (76)	209 (22)
Jun	1027 (232)	604 (67)	71 (4)
Aug	1053 (188)	1168 (76)	67 (27)
Nov	1134 (517)	2481 (175)	99 (16)
Bare			
Apr	346 (43)	228 (93)	67 (9)
Jun	251 (47)	248 (39)	29 (1)
Aug	182 (17)	983 (61)	44 (18)
Nov	305 (30)	2273 (288)	10 (2)

roots and rhizomes. For 2 of the 4 nitrification experiments, these samples had higher rates than bulk sediments surrounding plants. In the other experiments, rates associated with sediment scraped from the roots and rhizomes were the same or lower than those within the vegetated sediments. These results do not clearly define the spatial dimensions of plant influence on the activity of microbial community. Finer-scale sampling, as well as incubation of roots and rhizomes themselves, might resolve this question.

#### $\text{NH}_4^+$ production and plant demand

The depth distributions of ammonification within both *Potamogeton perfoliatus* and *Zostera marina* beds were very similar, although the magnitude and sea-



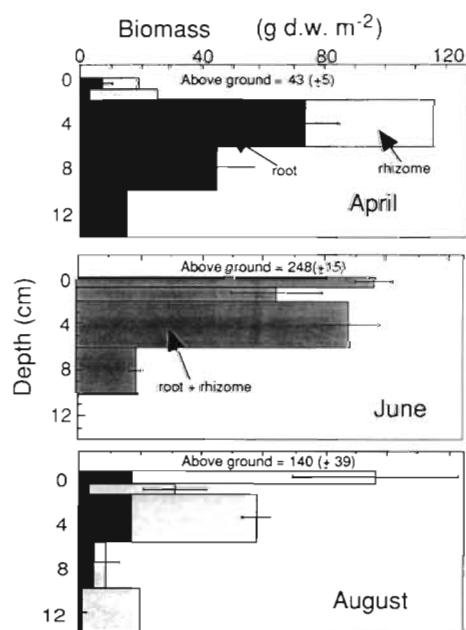


Fig. 7 *Zostera marina*. Root + rhizome biomass ( $\text{g m}^{-2}$ ) in (a) April, (b) June, and (c) August. Mean  $\pm$  standard errors ( $n = 3$ ) and biomass of aboveground plant parts are given

sonal pattern of ammonification were distinctly different. The highest ammonification rates (Table 1) coincided with the summer peak in productivity and biomass of *P. perfoliatus* (Kemp et al. 1984, Twilley et al. 1985). In contrast, the correspondence between *Z. marina* productivity and ammonification rates was not as strong, with ammonification rates slightly higher in April and November than in the summer (Table 2).

Different sources of organic matter contribute to ammonification rates, and the importance of any par-

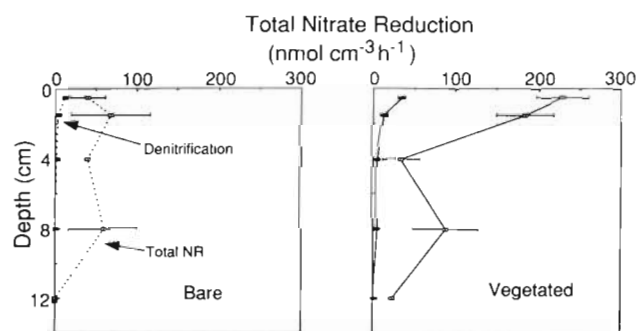


Fig. 8. Total nitrate reduction (open boxes) and denitrification (closed boxes) ( $\text{nmol cm}^{-3} \text{h}^{-1}$ ) in *Zostera marina* beds (continuous lines) and adjacent bare sediments (dotted lines) in April

ticular source may vary seasonally. Decomposition of allochthonous particulate organic nitrogen, such as from the spring phytoplankton bloom, would be most important in the early summer (Malone et al. 1988). Deamination of dissolved organic nitrogen released from macrophyte roots may represent a small fraction of ammonification, but release would be greatest when production is high (Smith et al. 1984a, Smith et al. 1988). A third source of organic matter is autochthonous biomass: roots, rhizomes, leaves and epiphytes (Kenworthy & Thayer 1984).  $\text{NH}_4^+$  production from mineralization of these tissues could occur throughout the year from turnover of leaves, roots and rhizomes, but peak rates would probably occur after the late summer senescent period. Assuming all senescent leaf biomass to be retained within the *Potamogeton perfoliatus* and *Zostera marina* beds,  $\text{NH}_4^+$  production through decomposition of plant biomass (Figs. 2 and 7) would contribute ca 20 % of the observed ammonifica-

Table 3. Ranges of rates of nitrogen transformation processes (ammonification [A], nitrification [N], denitrification [D] and  $\text{NO}_3$  reduction [NR]) from sediments occupied by *Zostera marina* and other submersed macrophytes,  $\text{nmol cm}^{-3} \text{h}^{-3}$

Macrophyte	A	N	D	NR	Source
<i>Zostera</i> spp.					
<i>Z. marina</i>	0–90	1–27	0–35	30–230	This study (vegetated)
<i>Z. marina</i>	6–160	7	0–3	–	Iizumi et al. (1980, 1982)
<i>Z. marina</i>	–	81	0–30	54	Koike & Hattori (1978a, b)
<i>Z. marina</i>	7–35	–	–	–	Dennison et al. (1987)
<i>Z. marina</i>	0–1	–	–	–	Short (1987)
<i>Z. marina</i>	0–13	–	–	–	Blackburn et al. (pers. comm.)
<i>Z. capricorni</i>	5–14	–	–	7–10	Boon et al. (1986a, b)
<i>Z. novaezelandica</i>	–	–	2–7	2–8	Kaspar (1983)
Other submersed macrophytes					
<i>Potamogeton perfoliatus</i>	0–70	0–450	0–60	81–260	This study (vegetated)
<i>Syringodium filiforme</i>	2–48	–	–	–	Short et al. (1985)
<i>Thalassia testudinum</i>	0–1	–	–	–	Kemp et al. (1988)
<i>Eriocaulon septangulare</i>	–	1–7	–	–	Kepkay (1985)
<i>Littorela uniflora</i>	–	–	0–2	–	Christensen & Sørensen (1986)

tion rates in these macrophyte beds. The actual contribution of decomposing *Z. marina* biomass to ammonification may be less since about 90% of the nitrogen in senescing leaves is translocated to other tissues (Borum et al. 1989). In addition, some of the leaf biomass is probably transported outside the beds by tides and currents. Further research is needed to resolve the contribution of allochthonous versus autochthonous material to ammonification.

$\text{NH}_4^+$  turnover in sediments with both *Potamogeton perfoliatus* and *Zostera marina* was high. Calculations of  $\text{NH}_4^+$  turnover, based on ammonification rates and porewater  $\text{NH}_4^+$  pools, in the top 8 cm, were from 1 to  $2 \text{ d}^{-1}$  in sediments with *P. perfoliatus*, and 4 to  $10 \text{ d}^{-1}$  in *Z. marina* sediments (Table 4). These turnover rates are faster than rates of about  $1.5 \text{ d}^{-1}$  reported for a shallow (1.3 m) *Z. marina* bed in Massachusetts, USA (Dennison et al. 1987). Enhanced abundances of macrofauna, associated with *Z. marina* sediments (Homziak et al. 1982), may contribute to the relatively increased  $\text{NH}_4^+$  pools and turnover rates resulting from animal excretion and bioturbation (Henriksen et al. 1980).

Ammonification within grassbeds represents a potentially important source of nitrogen for plant growth. During May and July in *Potamogeton perfoliatus* beds,  $\text{NH}_4^+$  concentrations were lower in vegetated than bare sediments (Fig. 2). This is not surprising when one compares ammonification and calculated plant uptake of nitrogen (Table 4). Ammonification rates were integrated over the top 8 cm to estimate an ambient rate of net ammonium production. We assumed that the rates from the slurry experiments are comparable to rates in intact sediments (as in Aller & Yingst 1980), since the sediments were incubated anaerobically without addition of any substrate (as in nitrification and denitrification experiments). In May,

when *P. perfoliatus* biomass was low, ammonification rates were slightly less than the calculated uptake rates. In July, plant uptake was about 10 times the ammonification rate, which led to a depletion of extractable  $\text{NH}_4^+$  concentrations in vegetated sediments, relative to bare sediments (Fig. 2). In September when productivity was reduced, ammonification rates exceeded plant demand. An important assumption of this calculation of plant nitrogen demand by *P. perfoliatus* is that remobilization and translocation of nitrogen from senescing leaves is small.

In contrast, extractable  $\text{NH}_4^+$  concentrations were usually higher in sediments vegetated with *Zostera marina* than adjacent bare sediments.  $\text{NH}_4^+$  produced during ammonification was in excess of the estimated nitrogen required by the plants (Table 4). In fact, throughout the growing season plant nitrogen demand was always less than the  $\text{NH}_4^+$  supplied by ammonification. This is similar to what both Kenworthy et al. (1982) and Dennison et al. (1987) reported for *Z. marina* beds in North Carolina and Massachusetts, respectively. In contrast, plant nitrogen demand exceeded regeneration when organic inputs were lower, where eelgrass was recolonizing an area (Short 1983) and in *Potamogeton perfoliatus* beds (Table 4).

### Nitrification

In coastal marine sediments, nitrification tends to be controlled by availability of both  $\text{NH}_4^+$  and  $\text{O}_2$  (Henriksen & Kemp 1988), although high concentrations of sulfide may be inhibitory (Hansen et al. 1981). Submersed plants directly affect nitrification by 2 processes: by uptake of  $\text{NH}_4^+$  that would otherwise be nitrified, and by root release of  $\text{O}_2$  which oxidizes the

Table 4. Comparison of rates of calculated plant nitrogen uptake with rates of sediment ammonification and porewater  $\text{NH}_4^+$  pools and turnover in top 8 cm of sediments containing *Potamogeton perfoliatus* and top 6 cm of sediments containing *Zostera marina*

Month	Productivity <sup>a</sup> ( $\text{mg O}_2 \text{ g}^{-1} \text{ d}^{-1}$ )	Biomass <sup>a</sup> ( $\text{g m}^{-2}$ )	Nitrogen demand <sup>b</sup> ( $\text{mmol N m}^{-2} \text{ d}^{-1}$ )	Ammonification ( $\text{mmol N m}^{-2} \text{ d}^{-1}$ )	$\text{NH}_4^+$ pool ( $\text{mmol m}^{-2}$ )	Turnover ( $\text{d}^{-1}$ )
<i>Potamogeton perfoliatus</i>						
May	49.0	50	3.7	2.2	2.6	0.8
July	104.4	467	60.2	6.3	6.2	1.0
September	3.1	174	0.7	2.9	1.4	2.1
<i>Zostera marina</i>						
April	29.2	44	1.5	10.3	1.9	5.4
June	12.0	250	2.4	22.8	5.2	4.4
August	5.6	324	1.4	21.3	2.0	10.6
November	12.2	183	2.6	8.5	1.7	5.0

<sup>a</sup> Net daytime apparent  $\text{O}_2$  productivity measured at ambient light with excised *P. perfoliatus* and *Z. marina* leaves incubated in BOD bottles (see text)

<sup>b</sup> Assuming C/N atomic ratios of 18.1 for *P. perfoliatus* (Twilley et al. 1986) and 20 for *Z. marina* (W. Dennison pers. comm.), and a metabolic O/C molar ratio of 1.2

rhizosphere, creating a more favorable environment for aerobic processes (Wium-Andersen & Andersen 1972, Jaynes & Carpenter 1986). In this study, potential nitrification rates in the sediments with *Potamogeton perfoliatus* were highest in July and about an order of magnitude greater than rates in *Zostera marina* beds. The relatively high rates of root  $O_2$  release from *P. perfoliatus* compared to *Z. marina* (Sand-Jensen et al. 1982, Smith et al. 1984b, Kemp & Murray 1986, Caffrey 1989) may account for the higher potential nitrification rates in *P. perfoliatus* beds. The rates of potential nitrification in the *Z. marina* beds in this study were comparable to those previously reported in Izembek Lagoon, Alaska (Koike & Hattori 1978a; Table 3).

In most coastal marine sediments, nitrification is not generally limited by  $NH_4^+$  supply, since nitrifiers saturate at  $NH_4^+$  concentrations which are low (50 to 100  $\mu M$ ) compared to porewater  $NH_4^+$  pools (Henriksen & Kemp 1988). However, in sediments where plant demand for  $NH_4^+$  is high compared to production, competition between nitrifiers and plants may occur, particularly in close proximity to the roots. This appears to be occurring in the *Potamogeton perfoliatus* sediments studied here, since nitrification was significantly related to both  $NH_4^+$  concentration and ammonification rates in vegetated sediments but not in bare sediments. In contrast, the absence of a significant correlation between ammonification and nitrification in sediments with *Zostera marina* indicates that plant-microbial competition for  $NH_4^+$  was unimportant in regulating nitrification.

### $NO_3^-$ reduction: losses versus conservation

Denitrification depends on  $NO_3^-$  and organic carbon availability and is inhibited by sulfides (Sørensen 1987). Rates of denitrification potential within the *Potamogeton perfoliatus* beds were very similar to rates measured in this study and previously in sediments with *Zostera* spp. (Koike & Hattori 1978b, Izumi et al. 1980, Kaspar 1983; Table 3), but higher than those reported for oligotrophic lake sediments vegetated with *Littorella uniflora* (Christensen & Sørensen 1986). These high rates of denitrification and  $NO_3^-$  reduction have been associated with plant roots (Blotnick et al. 1980, Christensen & Sørensen 1986). Although denitrification potential was enhanced in the sediments with *P. perfoliatus* compared to bare sediments, the rates did not exhibit the summer maximum that ammonification and nitrification potential did. Instead, denitrification was highest in spring when  $NO_3^-$  concentrations were high and insignificant in the summer when porewater  $NO_3^-$  concentrations were less than 1  $\mu M$ .

Denitrification potential within the *Zostera marina*

bed also exhibited a summer minimum and was strongly correlated to  $NO_3^-$  concentration. Since  $NO_3^-$  concentration and denitrification were positively correlated,  $NO_3^-$  availability and nitrification may be the dominant factors controlling denitrification. Another factor that may have influenced seasonality of denitrification in these grassbeds is increased root release of dissolved organic carbon (Christensen & Sørensen 1986). Presumably, this process was active in the spring and fall when *Z. marina* productivity was high. Elevated porewater sulfide concentrations, which were probably maximal in summer, could also have inhibited denitrification in these beds or interfered with the denitrification assay (acetylene block) method (Sørensen et al. 1980).

Reduction of  $NO_3^-$  to  $NH_4^+$  is another important pathway, which results in nitrogen being retained within the system as  $NH_4^+$ , instead of being lost as  $N_2$  through denitrification. The relative availabilities of  $NO_3^-$  and organic carbon appear to control which pathway, denitrification or  $NO_3^-$  reduction to  $NH_4^+$ , predominates. Denitrification is favored when  $NO_3^-$  concentrations are high and organic carbon is low, while  $NO_3^-$  reduction to  $NH_4^+$  predominates when the opposite conditions occur (Sørensen 1987, Koike & Sørensen 1988). These measurements may overestimate the importance of denitrification relative to  $NO_3^-$  reduction to  $NH_4^+$ , since sediment slurries were amended with high concentrations of  $NO_3^-$ .

Total  $NO_3^-$  reduction rates within *Potamogeton perfoliatus* beds were similar to those measured in *Zostera marina* beds in this study and by Koike & Hattori (1978a) (Table 3). In this study, denitrification comprised about 20 % of the total  $NO_3^-$  reduction in these sediments with *P. perfoliatus* and only 10 % in sediments with *Z. marina*. In sediments vegetated with *Zostera* spp., 2 patterns have been reported. In one case, denitrification dominated total  $NO_3^-$  reduction (Kaspar 1983); in the other denitrification was a small fraction of  $NO_3^-$  reduction (Boon et al. 1986b).  $NO_3^-$  reduction to  $NH_4^+$  represents a recycled source of  $NH_4^+$  which is capable of supporting both plant assimilation and bacterial nitrification.

### Balance among nitrogen transformation processes

An important characteristic of nitrogen cycling in the *Zostera marina* bed studied here was the relative balance among the measured nitrogen transformation processes. Vertically integrated ammonification rates and nitrification potentials were about the same order of magnitude, while denitrification potentials were usually about 10 % of nitrification (Table 2). This is in sharp contrast to rates in *Potamogeton perfoliatus* beds,

where nitrification potentials were usually 10 times greater than either ammonification or denitrification potentials (Table 1). However, nitrification and denitrification rates reported in this paper are potential rates of nitrogen transformations and represent the activity of the bacterial communities capable of using these specific metabolic pathways. For nitrifiers, which have a specialized metabolism (Fenchel & Blackburn 1979), high potentials indicate large bacterial populations and high nitrification rates, provided substrate availability and temperature are appropriate (Henriksen 1980). Bacteria which can use  $\text{NO}_3^-$  as a terminal electron acceptor are facultative denitrifiers also capable of various other metabolic pathways (Hattori 1983). For this reason, relations between potential and actual rates of denitrification are probably not as direct as for nitrification.

Supply of organic material, both dissolved and particulate, may be part of the reason why sediment nitrogen cycling processes differed markedly between the 2 study sites dominated by different plant species. Organic nitrogen and carbon are essential substrates for ammonification and denitrification, respectively. The *Potamogeton perfoliatus* beds, which are in ponds, are characterized by low inputs of allochthonous organic matter, so particulate organic inputs may have limited growth of ammonifier and denitrifier populations. In those sediments, not only were the different nitrogen transformations very closely coupled, but the competition between plant and bacteria communities for  $\text{NH}_4^+$  may have been more intense as well. In contrast, the microbial community in the *Zostera marina* bed was probably not limited by organic matter, so the different nitrogen transformations were not as closely related as they were in *P. perfoliatus* beds. Instead, other factors, such as temperature,  $\text{O}_2$  release from plant roots, and sulfide concentrations may have been more important in controlling nitrogen transformations.

In summary, the microbial communities responsible for key nitrogen transformations in sediments were enhanced by the presence and activities of both *Potamogeton perfoliatus* and *Zostera marina*: ammonification by inputs of organic nitrogen, nitrification by release of  $\text{O}_2$  by plant roots, and denitrification by production of  $\text{NO}_3^-$ . The magnitude of these effects varied on a seasonal basis and appeared to be related to the growth cycle of the 2 plant populations. While submersed plants affected nitrogen cycling through several mechanisms, the microbial community, in turn, regulated the nitrogen available to the plants for growth. These interactions between bacterial and plant communities create a complex network of controls which regulates nitrogen cycling within these 2 grass-beds.

**Acknowledgements.** We thank R. Allen, M. Mayer, C. Miller, L. Murray, J. Neundorfer, M. Owens, D. Pohle, R. T. Randall, W. T. Randall, P. Sampou and A. Silvia for their assistance in the field and laboratory and W. Boynton, W. Dennison, T. Fisher, and D. Rice for their review of an earlier version of this manuscript. Computer support was provided by the Computer Science Center, University of Maryland. Sigma-Xi (Grants in Aid of Research), National Science Foundation (BSR 8801259), Maryland Sea Grant College and Horn Point Environmental Laboratories provided support for this research.

#### LITERATURE CITED

- Aller, R., Yingst, J. (1980). Relationship between microbial distributions and the anaerobic decomposition of organic matter in surface sediments of Long Island Sound, USA. *Mar. Biol.* 56: 29–42
- American Public Health Association (APHA) (1985). Standard methods for the examination of water and wastewater, 16th edn. American Public Health Assoc., Washington, D.C.
- Blotnick, J. R., Rho, J., Gunner, H. B. (1980). Ecological characteristics of the rhizosphere microflora of *Myriophyllum heterophyllum*. *J. environ. Qual.* 9: 207–210
- Boon, P. I. (1986). Nitrogen pools in seagrass beds of *Cymodocea serrulata* and *Zostera capricorni* of Moreton Bay, Australia. *Aquat. Bot.* 25: 1–19
- Boon, P. I., Moriarty, D. J. W., Saffigna, P. G. (1986a). Rates of ammonium turnover and the role of amino-acid deamination in seagrass (*Zostera capricorni*) beds of Moreton Bay, Australia. *Mar. Biol.* 91: 259–268
- Boon, P. I., Moriarty, D. J. W., Saffigna, P. G. (1986b). Nitrate metabolism in sediments from seagrass (*Zostera capricorni*) beds of Moreton Bay, Australia. *Mar. Biol.* 91: 269–275
- Borum, J., Murray, L., Kemp, W. M. (1989). Aspects of nitrogen acquisition and conservation in eelgrass plants. *Aquat. Bot.* 35: 289–300
- Caffrey, J. M. (1989). The effect of submersed macrophytes on nitrogen cycling in estuarine sediments. Ph. D. dissertation, University of Maryland
- Capone, D. G. (1983).  $\text{N}_2$  fixation in seagrass communities. *J. mar. technol. Soc.* 17: 32–37
- Capone, D. G., Taylor, B. F. (1980). Microbial nitrogen cycling in a seagrass community. In: Kennedy, V. S. (ed.) *Estuarine perspectives*. Academic Press, New York, p. 153–161
- Capone, D. G., Penhale, P. A., Oremland, R. S., Taylor, B. F. (1979). Relationship between productivity and  $\text{N}_2$  ( $\text{C}_2\text{H}_2$ ) fixation in a *Thalassia testudinum* community. *Limnol. Oceanogr.* 24: 117–125
- Christensen, P. B., Sørensen, J. (1986). Temporal variation of denitrification activity in plant covered, littoral sediment (Lake Hampen, Denmark). *Appl. environ. Microbiol.* 51: 1174–1179
- Dennison, W. C., Alberte, R. S. (1985). Role of daily light period in the depth distribution of *Zostera marina* (eelgrass). *Mar. Ecol. Prog. Ser.* 25: 51–61
- Dennison, W. C., Aller, R. C., Alberte, R. S. (1987). Sediment ammonium availability and eelgrass (*Zostera marina*) growth. *Mar. Biol.* 94: 469–477
- Fenchel, T., Blackburn, T. H. (1979). *Bacteria and mineral cycling*. Academic Press, New York
- Fonseca, M. S., Fisher, J. S. (1986). A comparison of canopy friction and sediment movement between four species of seagrass with reference to their ecology and respiration. *Mar. Ecol. Prog. Ser.* 29: 15–22

- Fonseca, M. S., Fisher, J. S., Zieman, J. C., Thayer, G. W. (1982). Influence of the seagrass, *Zostera marina* L., on current flow. *Estuar. coast. Shelf Sci.* 15: 351–364
- Goldsborough, W. J., Kemp, W. M. (1988). Light responses of a submersed macrophyte: implications for survival in turbid tidal waters. *Ecology* 69: 1775–1786
- Hansen, J. I., Henriksen, K., Blackburn, T. H. (1981). Seasonal distribution of nitrifying bacteria and rates of nitrification in coastal marine sediments. *Microb. Ecol.* 7: 297–304
- Hattori, A. (1983). Denitrification and dissimilatory nitrate reduction. In: Carpenter, E. J., Capone, D. G. (eds.) *Nitrogen in the marine environment*. Academic Press, New York, p. 191–232
- Henriksen, K. (1980). Measurement of in situ rates of nitrification in sediment. *Microb. Ecol.* 6: 329–337
- Henriksen, K., Kemp, W. M. (1988). Nitrification in estuarine and coastal marine sediments: methods, patterns and regulating factors. In: Blackburn, T. H., Sørensen, J. (eds.) *Nitrogen cycling in coastal marine environments*. John Wiley and Sons, New York, p. 207–250
- Henriksen, K., Hansen, J. I., Blackburn, T. H. (1980). The influence of benthic infauna on exchange rates of inorganic nitrogen between sediment and water. *Ophelia suppl.* 1: 249–256
- Homziak, J., Fonseca, M. S., Kenworthy, W. J. (1982). Macro-benthic community structure in a transplanted eelgrass (*Zostera marina*) meadow. *Mar. Ecol. Prog. Ser.* 9: 211–221
- Iizumi, H., Hattori, A., McRoy, C. P. (1980). Nitrate and nitrite in interstitial waters of eelgrass beds in relation to the rhizosphere. *J. exp. mar. Biol. Ecol.* 47: 191–201
- Iizumi, H., Hattori, A., McRoy, C. P. (1982). Ammonium regeneration and assimilation in eelgrass (*Zostera marina*) beds. *Mar. Biol.* 66: 59–65
- Jaynes, M. L., Carpenter, S. R. (1986). Effects of vascular and nonvascular macrophytes on sediment redox and solute dynamics. *Ecology* 67: 875–882
- Jenkins, M. C., Kemp, W. M. (1984). The coupling of nitrification and denitrification in two estuarine sediments. *Limnol. Oceanogr.* 29: 608–619
- Jørgensen, N. O. G., Blackburn, T. H., Henriksen, K., Bay, D. (1981). The importance of *Posidonia oceanica* and *Cymodocea nodosa* as contributors of free amino acids in water and sediment of seagrass beds. *Mar. Ecol.* 2: 97–112
- Kaspar, H. F. (1983). Denitrification, nitrate reduction to ammonium, and inorganic nitrogen pools in intertidal sediments. *Mar. Biol.* 74: 133–139
- Kemp, W. M., Murray, L. (1986). Oxygen release from roots of the submerged macrophyte, *Potamogeton perfoliatus* L.: regulating factors and ecological implications. *Aquat. Bot.* 26: 271–283
- Kemp, W. M., Boynton, W. R., Twilley, R. R., Stevenson, J. C., Means, J. C. (1983). The decline of submerged vascular plants in upper Chesapeake Bay: summary of results concerning possible causes. *J. mar. technol. Soc.* 17: 78–89
- Kemp, W. M., Boynton, W. R., Twilley, R. R., Stevenson, J. C., Ward, L. G. (1984). Influences of submersed vascular plants on ecological processes in upper Chesapeake Bay. In: Kennedy, V. S. (ed.) *The estuary as a filter*. Academic Press, New York, p. 367–394
- Kemp, W. M., Boynton, W. R., Stevenson, J. C., Hopkinson, C. S., Jr, Day, J. W., Jr, Yanez-Arancibia, A. (1988). Ammonium regeneration in the sediments of a tropical seagrass bed (*Thalassia testudinum*) community, Terminos Lagoon. In: Yanez-Arancibia, A., Day, J. W. Jr (eds.) *Ecology of coastal ecosystems in the southern Gulf of Mexico: the Terminos lagoon region*. Universidad Nacional Autonoma de Mexico, Mexico, p. 181–192
- Kenworthy, W. J., Thayer, G. W. (1984). Production and decomposition of the roots and rhizomes of seagrasses, *Zostera marina* and *Thalassia testudinum*, in temperate and subtropical marine ecosystems. *Bull. mar. Sci.* 35: 364–379
- Kenworthy, W. J., Zieman, J. C., Thayer, G. W. (1982). Evidence for the influence of seagrasses on the benthic nitrogen cycle in a coastal plain estuary near Beaufort, North Carolina (USA). *Oecologia (Berl.)* 54: 152–158
- Kepkay, P. E. (1985). Microbial manganese oxidation and nitrification in relation to occurrence of macrophyte roots in a lacustrine sediment. *Hydrobiologia* 128: 135–142
- Kikuchi, T. (1980). Faunal relationships in temperate seagrass beds. In: Phillips, R. C., McRoy, C. P. (eds.) *Handbook of seagrass biology: an ecosystem perspective*. Garland, p. 153–172
- Koike, I., Hattori, A. (1978a). Simultaneous determinations of nitrification and nitrate reduction in coastal sediments by a  $^{15}\text{N}$  dilution technique. *Appl. environ. Microbiol.* 35: 853–857
- Koike, I., Hattori, A. (1978b). Denitrification and ammonia formation in anaerobic coastal sediments. *Appl. environ. Microbiol.* 35: 853–857
- Koike, I., Sørensen, J. (1988). Nitrate reduction and denitrification in marine sediments. In: Blackburn, T. H., Sørensen, J. (eds.) *Nitrogen cycling in coastal marine environments*. John Wiley and Sons, New York, p. 251–273
- Lipschultz, F., Cunningham, J. J., Stevenson, J. C. (1979). Nitrogen fixation associated with four species of submerged angiosperms in the central Chesapeake Bay. *Estuar. coast. mar. Sci.* 9: 813–818
- Malone, T. C., Crocker, L. H., Pike, S. E., Wendler, B. W. (1988). Influences of river flow on the dynamics of phytoplankton production in a partially stratified estuary. *Mar. Ecol. Prog. Ser.* 48: 235–249
- O'Neill, J. M., Capone, D. G. (1989). Nitrogenase activity in tropical carbonate marine sediments. *Mar. Ecol. Prog. Ser.* 56: 145–156
- Orth, R. J. (1977). The importance of sediment stability in seagrass communities. In: Coull, B. C. (ed.) *Ecology of marine benthos*. Univ. S. Carolina Press, Columbia, p. 281–300
- Rosenfeld, J. K. (1979). Ammonium adsorption in nearshore anoxic sediments. *Limnol. Oceanogr.* 24: 356–364
- Sand-Jensen, K., Prahl, C., Stockholm, H. (1982). Oxygen release from roots of submerged aquatic macrophytes. *Oikos* 38: 349–354
- Short, F. T. (1983). The responses of interstitial ammonium in eelgrass (*Zostera marina* L.) beds to environmental perturbations. *J. exp. mar. Biol. Ecol.* 68: 195–208
- Short, F. T. (1987). Effects of sediment nutrients on seagrasses: literature review and mesocosm experiment. *Aquat. Bot.* 27: 41–57
- Short, F. T., Davis, M. W., Gibson, R. A., Zimmermann, C. F. (1985). Evidence for phosphorus limitation in carbonate sediments of the seagrass *Syringodium filiforme*. *Estuar. coast. Shelf Sci.* 20: 419–430
- Smith, G. W., Hayasaka, S. S., Thayer, G. W. (1984a). Ammonification of amino acids by rhizoplane microflora of *Zostera marina* L. and *Halodule wrightii* Aschers. *Botanica mar.* 26: 23–27
- Smith, R. D., Dennison, W. C., Alberte, R. S. (1984b). Role of seagrass photosynthesis in root aerobic processes. *Plant Physiol.* 74: 1055–1058
- Smith, R. D., Pregall, A. M., Alberte, R. S. (1988). Effects of anaerobiosis on root metabolism of *Zostera marina* (eel-



- grass): implications for survival in reducing sediments. *Mar. Biol.* 98: 131–141
- Solorzano, L. (1969). Determination of ammonia in natural waters by the phenylhypochlorite method. *Limnol. Oceanogr.* 14: 799–801
- Sørensen, J. (1978). Denitrification rates in marine sediment as measured by the acetylene inhibition technique. *Appl. environ. Microbiol.* 36: 139–143
- Sørensen, J. (1987). Nitrate reduction in marine sediment: pathways and interactions with iron and sulfur cycling. *Geomicrobiol. J.* 5: 401–421
- Sørensen, J., Tiedje, J. M., Firestone, R. B. (1980). Inhibition by sulfide of nitric and nitrous oxide reduction by denitrifying *Pseudomonas fluorescens*. *Appl. environ. Microbiol.* 39: 105–108
- Stevenson, J. C. (1988). Comparative ecology of submersed grass beds in freshwater, estuarine and marine environments. *Limnol. Oceanogr.* 33: 867–893
- SYSTAT (1986). The system for statistics. SYSTAT, Inc. Evanston, IL.
- Twilley, R. R., Kemp, W. M., Staver, K. W., Stevenson, J. C., Boynton, W. R. (1985). Nutrient enrichment of estuarine submersed vascular plant communities. 1. Algal growth and effects on production of plants and associated communities. *Mar. Ecol. Prog. Ser.* 23: 179–191
- Twilley, R. R., Ejdung, G., Romare, P., Kemp, W. M. (1986). A comparative study of decomposition, oxygen consumption and nutrient release for selected aquatic plants occurring in an estuarine environment. *Oikos* 47: 190–198
- Ward, L. G., Kemp, W. M., Boynton, W. R. (1984). The influence of waves and seagrass communities on suspended sediment dynamics in an estuarine embayment. *Mar. Geol.* 59: 85–103
- Wetzel, R. L., Penhale, P. A. (1983). Production ecology of seagrass communities in the lower Chesapeake Bay. *J. mar. technol. Soc.* 17: 22–31
- Wium-Andersen, S., Andersen, J. M. (1972). The influence of vegetation on the redox profile of the sediment of Grane Langso, a Danish *Lobelia* lake. *Limnol. Oceanogr.* 17: 948–952

*This article was presented by Dr G. W. Thayer, Beaufort, N. Carolina, USA*

*Manuscript first received: June 20, 1989  
Revised version accepted: June 8, 1990*