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Nitrogen biogeochemistry in an unpolluted estuary: the importance of benthic denitrification

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ABSTRACT: Denitrification rates (N_2 production), and benthic ammonium, nitrate and oxygen fluxes were measured along a salinity gradient in Ochlockonee Bay (Florida, USA) over an annual cycle. Denitrification rates ranged from 0 to 210 µg-at N m^{-2} h^{-1}. Lowest rates of denitrification were measured in late winter and highest rates in late spring. A major portion of the organic nitrogen cycled through the sediments is denitrified; on an annual basis an average of 69 % of the benthic nitrogen flux is N_2 and 31 % is ammonia, nitrite and nitrate. During summer the amount of nitrogen lost from the recycling pathway by denitrification is equivalent to about 40 % of the nitrogen requirements of the phytoplankton. The major source of NO_3^- or NO_2^- for denitrification is from nitrification in the sediments, not diffusion of NO_3^- or NO_2^- from the overlying water. Denitrification is a major removal mechanism for nitrogen in Ochlockonee Bay, and removes, on an annual basis, an amount of nitrogen equivalent to 54 % of the river input of dissolved inorganic nitrogen. N_2O fluxes were small compared to N_2 fluxes; N_2O/N_2 ratios in March were less than 0.01.

INTRODUCTION

It is well recognized that external ('new') inputs of nitrogen to most estuaries are not sufficient to supply the nitrogen needed to support annual primary production rates; rapid *in situ* recycling of nitrogen supplies a major portion of the nitrogen used by the algae (Harrison & Hobbie 1974, reviews by Nixon 1981, Boynton et al. 1982). Both the water column (Harrison 1978, Caperon et al. 1979, Glibert 1982) and the sediments (Rowe et al. 1975, Nixon et al. 1976, Blackburn & Henriksen 1983 and others) are important sites for nitrogen recycling in estuaries and other shallow coastal systems. However, during the mineralization of organic matter in sediments and in low oxic water, nitrogen can be lost from the recycling pathway as N_2 if denitrification is occurring. The extent to which denitrification removes nitrogen as it is mineralized in a system influences the amount of nitrogen available for primary production.

Rates of denitrification have now been measured in sediments from numerous estuaries and other coastal marine systems (see Seitzinger [in press] for a review). However, only a few studies have evaluated the quantitative importance of denitrification relative to other nitrogen fluxes in coastal sediments (Billen 1978, Seitzinger et al. 1984, Kaspar et al. 1985). Denitrification in estuaries can also be important as a sink for anthropogenic and continentally derived N inputs to the oceans (Seitzinger et al. 1984). However, there is only one estuary where sufficient data have been reported to evaluate this (Seitzinger et al. 1984).

Most studies of estuarine nutrient dynamics, including essentially all estuarine denitrification studies, have been carried out in estuaries that receive significant amounts of pollutants, including nutrients from sewage sources. The purpose of the present study was to examine the role of the benthos as a sink for nitrogen in an estuary that is not dominated by anthropogenic nutrient inputs and pollutants. The magnitude of denitrification along a salinity gradient relative to benthic NH_4^+ and NO_3^- fluxes is addressed as well as the importance of denitrification in decreasing the transport of continentally derived nitrogen to the oceans.

METHODS

Study site. Ochlockonee Bay is located on the 'panhandle' of Florida and empties into Appalachia Bay and the Gulf of Mexico (Fig. 1). The major freshwater flow into the bay is from the Ochlockonee River. The Apalachicola National Forest and St. Mark's National Wildlife refuge comprise the lower 105 km of the river's relatively undisturbed watershed. The upper

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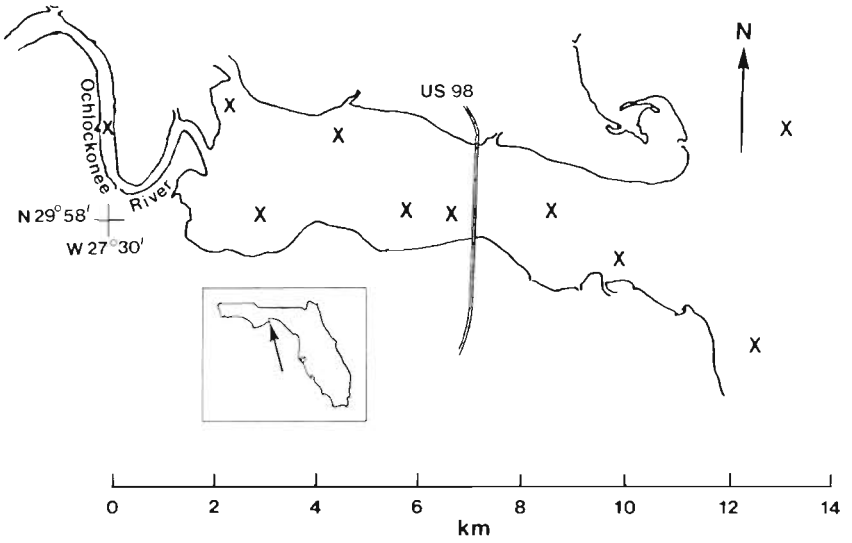

Fig. 1 Location of sediment sampling sites in Ochlockonee Bay (Florida, USA) for denitrification and sediment-water nutrient flux measurements. Distances (km) of sites from river site are indicated

watershed is forested and sparsely populated. We know of no sewage treatment plants or industries that discharge into the river. The estuary is approximately 10 km long by 2.4 km wide. Average tidal range is 0.6 m and mean depth at low tide is 1 m (Kaul & Froelich 1984). The depth of the lower 5 km of the Ochlockonee River ranges from 3 to 8 m. The upper half of the bay is generally the shallowest, and much of the lower half of the bay ranges from 2 to 3 m deep at low tide. The water column is well-mixed with vertical salinity gradients of less than 1 ppt $(m\text{-}depth)^{-1}$. Phytoplankton production during summer is approximately 26 to 31 mg C m^{-2} h^{-1} (based on a 12 h day) as estimated from ^{14}C productivities (Myers & Iverson 1981) and from modelling nutrient distributions in the bay (Kaul & Froelich 1984). Nutrient concentrations are low throughout the bay, and generally decrease from the head of the bay, where the Ochlockonee River enters, to the mouth of the bay. Typical nutrient concentration ranges at the head and at the mouth of the bay, respectively, are: ammonia 1.8 to 2.5 μM and <0.5 to 1.5 μM; nitrate 3 to 8 μM and <0.1 to 1.8 μM; phosphate 0.5 to 1.6 μM and 0.01 to 0.4 μM (Kaul & Froelich 1984, Seitzinger unpubl.).

Denitrification and sediment–water nutrient flux measurements. An initial study was carried out in June 1984 to assess the spatial heterogeneity of benthic nutrient fluxes in Ochlockonee Bay (Seitzinger unpubl.). The results of that study were used to choose a limited number of sites, representative of larger areas of the bay, for measurements of denitrification and benthic nutrient fluxes over an annual cycle. Sediment–water fluxes of ammonium, nitrite plus nitrate, phosphate and oxygen were made at 30 locations throughout the bay (1 core per site) between 4 and 9 Jun 1984. The spatial variability in the sediment–water nutrient and oxygen fluxes was low (Seitzinger unpubl.). Therefore, sites for seasonal measurements of denitrification and benthic nutrient fluxes were chosen that spanned the length of the estuary and included locations in the river, upper, mid and lower bay and outside the mouth of the bay (Fig. 1).

Sediment cores for denitrification and benthic nutrient flux measurements were collected on 21 Apr 1984, 4 to 5 Jun 1984, 17 Nov 1984, and 19 Mar 1985 from 5 areas in the bay. The sediment cores (6.7 cm diameter, ca 15 cm deep) were collected by SCUBA-equipped divers using plastic coring tubes. Care was taken during coring to avoid disturbance of the sediment surface and loss of flocculent material. At the time of sediment collection water was collected from each location in acid-washed carboys. The cores were kept dark and maintained at ambient bay water temperature during transport to the laboratory. Denitrification rates (N_2 production) and benthic nutrient fluxes were measured according to techniques previously described in detail in Seitzinger et al. (1980) and Seitzinger & Nixon (1985); only the general outline and recent modifications of the method are described here.

The sediment cores (7 cm deep; 1 core per sampling location) were transferred to gas-tight glass incubation chambers which had been filled with helium for a number of days prior to the beginning of the experiment. The cores were incubated in the dark with unfiltered bay water (~300 ml) which was stirred continuously with a floating magnetic stirring bar to facilitate the equilibration of dissolved gases with the overlying gas phase (~70 ml). Water over the cores was changed at approximately 24 to 48 h intervals with freshly prepared low-N_2 bay water obtained by flushing water from the site of sediment collection with a gas mixture of 21 % O_2 and the balance He. The incubation temper-

ature was the same as the ambient bay water temperature at the time of sediment collection. The oxygen concentration in the water over the sediments throughout the experiment was always above 70 % saturation relative to atmospheric O_2.

Duplicate samples (100 µl) of the gas phase were taken for N_2 and O_2 analysis from each chamber approximately 6, 24, and occasionally 48 h after the water was changed. Net N_2 and O_2 fluxes across the sediment–water interface were calculated knowing the changes in concentration between sequential samples during an incubation and the volume of the gas phase in each chamber. Gas samples were injected directly into a gas chromatograph (Shimadzu, Model GC-8A) equipped with a thermal conductivity detector (2 m × 0.318 cm o.d. stainless steel columns packed with 45/60 mesh Molecular Sieve 5A, He carrier gas flow rate 25 cm^{-3} min^{-1}).

Previous experiments (Seitzinger 1982) showed that the N_2 initially dissolved in the pore waters became equilibrated with the low-N_2 overlying water in about 10 d. Ten d also appears to be sufficient in the present experiments because N_2 fluxes were below the level of detection (<25 µg-at N m^{-2} h^{-1}) after 10 d from sediments with no measurable denitrification (see 'Results'), and the rate of N_2 flux did not decrease after 10 d from cores with measurable denitrification rates. Therefore, measurements made after 10 d of incubation with low-N_2 water were used for calculations of denitrification rates. Two to 4 separate denitrification rate (N_2 flux) measurements were made on each core.

Sediment–water nutrient fluxes were measured from the same cores used for denitrification measurements. Initial water samples for sediment–water nutrient fluxes were taken from a chamber after the water was changed over a core but before the chamber was closed for N_2 flux measurements. Final samples were taken approximately 24 or 48 h later after the final gas sample was collected. A portion of each sample was filtered through pre-rinsed glass fiber filters (Whatman 934-AH) and frozen for later analysis of nitrite plus nitrate (Technicon Industrial Systems 1977). The unfiltered portion of the sample was analyzed immediately for ammonium (Solorzano 1969). Ammonium samples were not filtered because comparisons of filtered and unfiltered samples consistently indicated a small contamination due to filtering.

Changes in nutrient concentrations attributable to water column processes were measured by incubating water from each station without sediment. These controls were treated and sampled using the same procedures as were used for sampling the water over the cores. The sediment–water flux of nutrients was calculated based on the volume of water over the cores and the change in nutrient concentration between initial and final water samples from the cores after correcting for any changes noted in the controls.

During the March experiment time series samples (1.5 ml) of the gas phase were taken for N_2O analysis from each chamber. Nitrous oxide fluxes were measured on days when N_2 fluxes were not. Samples were injected directly into a gas chromatograph (Shimadzu, Model GC-8A) equipped with a ^{63}Ni electron capture detector (340 °C) and a 4.3 m × 0.318 cm (o.d.) stainless steel column packed with Poropak Q, 80/100 mesh (CH_4/Ar carrier gas flow 20 cm^{-3} min^{-1}). The total amount of nitrous oxide in the water and gas phases of the chambers was calculated using the N_2O solubility equation of Weiss & Price (1980) for the appropriate temperature and salinity.

Controls. To demonstrate the gas tightness of the incubation chambers, the ability to sample from the chambers without contamination, and the biological (denitrification) source of the N_2 produced, the following measurements were made: (1) The N_2 concentration was measured over time in empty, helium-filled chambers. (2) The N_2 flux from the sediments was measured with anoxic conditions in the water and gas phases at the end of the November 1984 denitrification measurements.

Pore water ammonium concentrations were measured in an upper bay (2.5 km site) and a mid bay (5.4 km site) core in June 1985. Cores were sectioned at 1 cm intervals in a glove bag continuously flushed with N_2. Pore waters were extracted in the glove bag by filtering the wet sediment through pre-rinsed, dried, glass fiber filters. The samples were analyzed immediately for ammonium after dilution with deionized distilled water (Solorzano 1969).

Dissolved inorganic nitrogen inputs to the bay from the Ochlockonee River were estimated from the ammonium and nitrite plus nitrate concentrations measured in the river (<1 ‰ salinity). Water samples were collected the day after the sediment cores were collected. Samples for nitrite plus nitrate (Technicon Industrial Systems 1977) were filtered through pre-rinsed glass fiber filters and frozen for later analysis. Unfiltered ammonium samples were analyzed immediately (Solorzano 1969).

RESULTS

Measurable rates of denitrification (>25 µg-at N m^{-2} h^{-1}) were found in Ochlockonee Bay at all seasons (Table 1). Denitrification rates were lowest in late winter (March), ranging from 0 to 65 µg-at N m^{-2} h^{-1}, and highest in late spring (April), ranging from 100 to 210 µg-at N m^{-2} h^{-1}. No measurable denitrification was

Table 1. Denitrification rates (N_2 production), oxygen consumption, and net flux of $NO_3^- + NO_2^-$, NH_4^+, N_2O from intact sediment cores collected from Ochlockonee Bay, Florida, USA (see Fig. 1). Rates are mean rate for a core; units: µg-at (N or O) m^{-2} h^{-1}

Month	Distance (km)	Approx. depth (m)	Salinity (‰)	Temp. (°C)	N_2	NO_2+NO_3	NH_4	O_2 uptake	N_2O	Visual sediment characteristics*
Apr 84	0.0	2.0	0	21	155	4	2	1625		Mud, no animals visible
	2.4	1.3	0		130	7	16	1345		Medium sand, no animals visible
	4.4	1.0	0		100	4	4	1250		Find sand with mud, NT
	8.7	2.0	2		210	20	− 61	1840		Mud, very soft, NT
	12.1	3.3	6		155	0	105	1250		Medium sand with mud, MT
Jun 84	0.0	2.0	0	25	75	14	46	510		Mud, no animals visible
	2.9	1.5	0		35	2	63	1230		Mud, soft, FT
	5.8	2.0	1		105	28	83	1235		Mud, MT
	9.8	1.5	14		95	14	78	1360		Mud, no animals visible
	12.1	3.3	27		55	18	15	1120		Fine sand w/mud, no animals visible
Nov 84	2.4	1.5	8	16	110	14	− 6	925		Mud, soft, MT
	2.9	1.5	14		95	17	− 6	915		Mud, soft, FT
	6.6	2.0	14		130	49	−12	1045		Mud, very soft, FT
	8.7	2.0	18		80	20	− 2	615		Mud, soft, no animals visible
	11.8	1.3	2		0	0	5	400		Fine sand, no animals visible
Mar 85	0.0	1.0	0	17	30	10	6	510	0.04	Top 1 mm mud, below fine sand w/mud
	2.9	1.5	4		55	29	8	1590	0.05	Mud, soft, MT
	6.6	2.0	15		65	29	2	990	0.02	Mud, 1 oyster shell
	8.7	2.0	15		nm	18	2	1565	0.04	Mud, soft, MT
	12.1	3.3	22		0	22	26	2020	0.03	Fine sand, brittle stars

* FT: < 10 animal tubes per core; NT: 10 to 20 animal tubes per core; MT: > 20 animal tubes per core; nm: not measured

found in November or March in sediments collected outside the mouth of the bay. Rates of denitrification measured in June were similar to those measured in late fall. Within a season, there was no obvious relationship between denitrification rate and location within the bay, salinity, sediment characteristics or abundance of infauna (Table 1).

Sediment-water ammonium fluxes were generally low in April, November, and March (Table 1). At 4 locations in November and 1 in April there was a net flux of ammonium into the sediments. Ammonium fluxes were higher in June. There was no obvious pattern of ammonium fluxes with station location, temperature or salinity. There was a net flux of nitrite plus nitrate out of the sediments at all locations during all sampling times, except from sediments collected outside the mouth of the bay. In April and November sediments from that location exhibited no net flux of nitrite plus nitrate into or out of the sediments.

Pore water NH_4^+ concentrations increased with depth in cores collected from an upper and a mid-bay location in June 1985. At the upper bay location, ammonium concentrations increased from 90 µM in the top 0 to 1 cm to 320 µM in the 4 to 5 cm section. At the mid-bay location, ammonium concentrations increased from 72 µM in the top 0 to 1 cm to 190 µM in the 4 to 5 cm section.

Nitrous oxide fluxes were small relative to N_2 fluxes and ranged from 0.02 to 0.05 µg-at N m^{-2} h^{-1} in March (Table 1).

No N_2 influx into empty, helium-flushed incubation chambers could be detected (Table 2). At the end of the November denitrification measurements, the chambers were allowed to go anoxic and N_2 flux measurements

Table 2. N_2 production rates in empty helium-flushed chambers and in chambers with sediment cores during the November denitrification experiment with aerobic overlying water and with anaerobic overlying water. Rates reported as average flux (µg-at N m^{-2} h^{-1}) ± SD

Chamber #/Station	N_2 flux	No. measurements
Empty chambers		
#1	0	2
#2	0	2
Aerobic with sediments		
1.9 km	108 ± 59	4
2.5 km	95 ± 35	4
5.4 km	131 ± 59	4
7.2 km	79 ± 44	3
Anaerobic with sediments		
1.9 km	−6 ± 26*	6
2.5 km	−1 ± 31*	5
5.4 km	3 ± 17*	3
7.2 km	8 ± 12*	7

* Flux not different from zero ($\alpha = 0.05$)

were continued. As expected, N_2 fluxes under anoxic conditions were not significantly different from zero ($\alpha < 0.05$). Without oxygen no further nitrification could occur and thus no denitrification occurred.

Nitrogen inputs to Ochlockonee Bay from the Ochlockonee River during 1984–85 were estimated from measured $NH_4^+ + NO_3^- + NO_2^-$ concentrations in the river and by making certain adjustments to Kaul & Froelich's (1984) estimate of nitrogen inputs to the bay during 1980–81. Their estimate of 0.2175 moles N s^{-1} was made by multiplying the Ochlockonee River flow during 1980–81 by the nitrite plus nitrate concentration measured monthly in the river. I have made adjustments to their estimate for (1) the difference in river flow in 1980–81, a severe drought year, to that in 1984–85, a more normal flow year, and (2) the ammonium supplied to the bay from the river.

The river flow into Ochlockonee Bay varies substantially from year to year; the 23 yr average flow of the Ochlockonee River at the Jackson Bluff dam from 1960 through 1982 was 53 ± 24 m^3 s^{-1} (U.S. Geological Survey unpubl.). The average flow during the period of Kaul & Froelich's study (1980–1981) was 20 m^3 s^{-1} while during 1984–1985 it was approximately 3 times larger, 62 m^3 s^{-1} (U.S. Geological Survey unpubl.) and similar to the 23 yr average flow. The Jackson Bluff dam is about 105 km upstream from the mouth of the Ochlockonee River. The river flow at the dam is taken to be representative of the discharge into the bay because there are no major tributaries or distributaries between the dam and the mouth of the river.

Ammonium concentrations measured in the Ochlockonee River during 1984–85 ranged from 1.8 to 2.5 µM and averaged 38 % of the nitrite plus nitrate concentrations. These ammonium concentrations are similar to those measured in unpolluted tropical rivers reviewed by Meybeck (1982) which range from 0.5 to 2.9 µM. The river concentrations of nitrite plus nitrate (3 to 8 µM) measured during the present study were essentially the same as those measured during similar months in 1980–1981 by Kaul & Froelich (1984), despite the difference in river flow rates. Since the frequency of river nutrient concentration measurements made in the present study is low compared to the monthly measurements of Kaul & Froelich, adjustments to account for the ammonium inputs to Ochlockonee Bay were made by multiplying Kaul & Froelich's (1984) nitrite plus nitrate input rate by 1.38. The combined adjustments made to Kaul & Froelich's (1984) 1980–81 estimate of riverine N inputs to calculate the riverine inputs of inorganic N to Ochlockonee Bay in 1984–85 are: (0.2175 moles N s^{-1}) × 1.38 (ammonium) × 3 (flow) = 0.90 moles N s^{-1} or 135 µg-at N m^{-2} h^{-1} (area of bay 24×10^6 m^2; determined from planimetry of U.S. Geological Survey maps 00624 and 01142).

DISCUSSION

Denitrification removes nitrogen from a system by reducing nitrite or nitrate to N_2 gas. Three sources of nitrite and nitrate for sediment denitrification are: (1) $NO_3^- + NO_2^-$ diffusing into the sediments from the water column, (2) nitrification occurring in the sediments, and (3) $NO_3^- + NO_2^-$ advected through the sediments from groundwater inputs. No direct measurements of the latter are available in Ochlockonee Bay. However, it is not likely that groundwater is a major source of $NO_3^- + NO_2^-$ for denitrification in Ochlockonee Bay sediments since $NO_3^- + NO_2^-$ concentrations in the local groundwater are low and the sediments were isolated during the denitrification measurements from any new groundwater inputs.

The major source of nitrite and nitrate for denitrification in Ochlockonee Bay sediments appears to be nitrification in the sediments, not diffusion of nitrite or nitrate from the water column. This conclusion is based on the finding that there was no measurable flux of nitrite or nitrate from the overlying water into the sediments in which denitrification was occurring (Table 1).

Nitrification in the sediments appears to be the major source of $NO_3^- + NO_2^-$ for denitrification in most coastal marine sediments with oxic bottom waters (Seitzinger in press). This conclusion is based on the observation that there is generally a net flux of $NO_3^- + NO_2^-$ out of sediments in which denitrification is occurring, or the rate of uptake of $NO_3^- + NO_2^-$ from the water column is much less than that required to supply the reported denitrification rates (Table 3). Tama Estuary, Japan, sediments are an exception to this; there nitrate from the overlying water is the major source of nitrate for denitrification (Nishio et al. 1983). Nitrification in the sediments also appears to be the major source of nitrate for denitrification in a number of lakes and rivers (Seitzinger in press).

A large percentage of the $NO_3^- + NO_2^-$ produced in Ochlockonee Bay sediments is denitrified, as indicated by a comparison of the denitrification rates and the flux of $NO_3^- + NO_2^-$ out of the sediments (Table 1). An average (for all cores) of 80 % of the $NO_3^- + NO_2^-$ produced in the sediments is denitrified. (The production of $NO_3^- + NO_2^-$ was calculated as the sum of the measured $NO_3^- + NO_2^-$ flux out of the sediment plus the NO_3^- or NO_2^- required to supply the measured denitrification [Table 4].) This assumes no reduction of NO_3 or NO_2 to NH_4 (Koike & Hattori 1978, Sorensen 1978, Jenkins & Kemp 1984) and thus may be a maximum estimate. This efficiency of removal implies a tight coupling between nitrification and denitrification, which may be due to denitrification occurring in reduced microenvironments within the aerobic surface

Table 3. Sediment-water fluxes of ammonium, nitrate plus nitrite, and N_2, and percent of N flux due to denitrification (N_2 flux/[NH_4+NO_3+NO_2+N_2] flux) in coastal marine sediments. units: μg-at N m^{-2} h^{-1}

Location	Flux (μg-at N m^{-2} h^{-1}) NH_4	NO_3+NO_2	N_2	Percent N_2	Reference
Narragansett Bay, USA	91	10	59	37	Seitzinger et al. 1984
North Sea, Belgian Coast					
Coastal	73	98	49	22	Billen 1978
Offshore	41	49	16	15	Billen 1978
North Sea					
Belgian Coast	92	100	88	31	Vanderborght et al. 1977
Patuxent Estuary, USA	467	−42	133	20	Reported in Henriksen & Kemp in press
Tejo Estuary, Portugal					
Upper bay	28	212	172	42	Seitzinger unpubl. data
S. Island West Coast, New Zealand					
Stn R200	17	17	10	23	Kaspar et al. 1985
Stn R212	17	8	77	75	Kaspar et al. 1985
Stn 213	21	25	53	54	Kaspar et al. 1985
Lake Grevelingen, Netherlands					
Saltwater 1977		139•	29	21	de Vries & Hopstaken 1984
Ochlockonee Bay	15	18	73	69	This study
Tama Estuary, Japan					
December		− 28••	15		Nishio et al. 1983
May		−119••	59		Nishio et al. 1983

• Combined flux of NH_4 and NO_3+NO_2 reported; •• only NO_3+NO_2 flux reported

Table 4. Calculated sediment nitrification rates, percent of N efflux that is N_2, and percent of benthic oxygen consumption used for nitrification in Ochlockonee Bay sediments. Rates used for calculations are from Table 1

Month	Distance (km)	Sediment nitrification•	% O_2 consumption due to nitrification••	% Benthic N efflux that is N_2•••
Apr 84	0.0	159	39	96
	2.4	137	41	85
	4.4	104	33	93
	8.7	230	50	100
	12.1	155	50	60
Jun 84	0.0	89	70	55
	2.9	37	12	35
	5.8	133	43	49
	9.8	109	32	51
	12.1	73	26	63
Nov 84	2.4	124	54	93
	2.9	112	49	90
	6.6	179	69	78
	8.7	100	65	82
	11.8	0	0	0
Mar 85	0.0	40	31	65
	2.9	84	21	60
	6.6	94	38	68
	8.7			
	12.1	22	4	0

• Calculated as the sum of the measured NO_2+NO_3 flux out of the sediment plus the NO_3 that is required to supply the measured denitrification

•• Consumption of oxygen due to nitrification calculated, assuming only NO_3 produced, as nitrification rate (μg-at N m^{-2} h^{-1}) × 4 μg-at O used per μg-at NO_3 produced

••• (N_2 flux/[NH_4+NO_2+NO_3+N_2 flux]) × 100

sediments or within aerobic sediments lining the burrows of certain infauna (Henriksen et al. 1983). The high percentage of nitrate and nitrite produced in the sediments that is denitrified suggests that the nitrification rate is an important factor controlling the denitrification rate. Jenkins & Kemp (1984) observed a tight coupling between sediment nitrification and denitrification in Patuxent River (Chesapeake Bay, USA) estuary sediments where over 99 % of $^{15}N\text{-}NO_3^-$ produced in the sediments from added $^{15}N\text{-}NH_4^+$ was reduced to $^{15}N\text{-}N_2$. High efficiencies of nitrite and nitrate reduction by denitrification have also been reported in Narragansett Bay (Seitzinger et al. 1984) and from a MERL mesocosm nutrient-enrichment experiment (Seitzinger & Nixon 1985). Denitrification reduced 62 to 100 % of the nitrite and nitrate produced in those sediments; no pattern relative to temperature or degree of eutrophication was evident. Christensen & Rowe (1984) calculated that sediment-related nitrification supplied all the nitrate for denitrification in Gulf of Maine sediments, and that 26 % of the nitrate produced in the sediments was denitrified.

A major portion of the nitrogen cycled through the sediments in Ochlockonee Bay was not returned to the water column as ammonium, nitrite or nitrate for further use by the algae, but rather was lost from the system as N_2. As an annual average, 69 % of the benthic N flux ($NH_4^+ + NO_2^- + NO_3^- + N_2$) was N_2. (This was calculated from the average annual N fluxes estimated by plotting the average NH_4^+, $NO_2^- + NO_3^-$, or N_2 flux in the bay as a function of the month of sediment collection and integrating the area under the curve.) This is similar to the percentage of benthic N fluxes that is N_2 reported for other coastal marine sediments (Table 3). During summer in Ochlockonee Bay the amount of N lost from the benthic recycling pathway via denitrification is equivalent to approximately 40 % of the N requirements of the phytoplankton. This calculation is based on a summer primary production rate of 15 mg C m^{-2} h^{-1} averaged over 24 h (30 mg C m^{-2} h^{-1} for a 12 h day; Meyers & Iverson 1981, Kaul & Froelich 1984), a C:N ratio of the phytoplankton of 6.625 by atoms, and an average denitrification rate in June for the 5 locations of 73 µg-at N m^{-2} h^{-1}.

Benthic NH_4^+ fluxes are quantitatively greater than benthic $NO_3^- + NO_2^-$ fluxes in many coastal sediments (Nixon et al. 1976, Fisher et al. 1982, Boynton & Kemp 1985). As such, the magnitude and importance of nitrification in the sediments has been somewhat obscured. The slow rates of nitrification reported in coastal waters (Hattori et al. 1978, McCarthy et al. 1984) and rapid pelagic ammonification rates (Harrison 1978, Caperon et al. 1979, Glibert 1982) have also, I think, influenced our ideas about the relative rates of these 2 processes in sediments. However, as discussed above, a large percent of the nitrogen cycled through coastal sediments is denitrified and the $NO_3^- + NO_2^-$ to support that denitrification comes from nitrification in the sediments. A comparison of the magnitude of $NO_3^- + NO_2^- + N_2$ fluxes with NH_4^+ fluxes indicates that in Ochlockonee Bay and in other coastal sediments over half of the organic nitrogen cycled through the sediments is oxidized in the sediments to NO_3^- or NO_2^- (Table 3). In Ochlockonee Bay approximately 80 % of the nitrogen cycled through the sediments is nitrified. This calculation does not include the possible reduction of NO_3^- or NO_2^- to ammonium in the sediments (Koike & Hattori 1978, Sorensen 1978, Jenkins & Kemp 1984) and as such may be a minimum estimate.

It is interesting to compare rates of nitrification measured in coastal sediments with pelagic nitrification rates. In a review of nitrification in the marine environment, Kaplan (1983) reported that nitrification rates measured in coastal waters range from 0.0006 to 0.1 µmol N l^{-1} h^{-1}. However, rates of nitrification per unit volume in coastal sediments are generally several orders of magnitude greater and range from 0 to 0.05 µmol N cm^{-3} h^{-1} with rates commonly 0.02 µmol N cm^{-3} h^{-1} (Kaplan 1983). Assuming a sediment porosity of 50 %, sediment nitrification rates range from 0 to 100 µmol N l^{-1} h^{-1}, with 40 µmol N l^{-1} h^{-1} common. The high rates of nitrification in sediments relative to the water column may reflect the high NH_4^+ concentrations typical of coastal sediments. Of course a comparison of the total amount of $NO_3^- + NO_2^-$ produced in the water and sediments must take into consideration the total volume of the water column and the sediments in which nitrification occurs.

Nitrification alone in the sediments accounts for an average of 38 % of the sediment oxygen consumption (Table 4; average of all cores). (The oxygen consumption due to nitrification was calculated by multiplying the calculated nitrification rate [$NO_3^- + NO_2^-$ flux + N_2 production] for a core by 4, because 4 µg-at O_2-O are consumed per µg-at of NO_3^- produced.) Other studies have shown that nitrification accounts for a similarly high percentage of benthic oxygen consumption. In Narragansett Bay (Rhode Island, USA) approximately 30 to 50 % of the sediment oxygen consumption at the mid to lower bay sites was attributed to nitrification (Seitzinger et al. 1984) and 30 to 50 % across a eutrophication gradient in the MERL mesocosms (Seitzinger & Nixon 1985). Christensen & Rowe (1984) estimated that 35 % of the oxygen consumption in Gulf of Maine sediments is related to nitrification. In Danish coastal sediments and Chesapeake Bay sediments, nitrification accounts for a smaller percent of the sediment oxygen consumption, averaging 7 and 8 %, respectively (Henriksen & Kemp in press).

Denitrification rates were significantly ($\alpha = 0.01$) cor-

related with sediment oxygen consumption: N_2 (μg-at N m^{-2} h^{-1}) = 0.083 O_2 (μg-at O m^{-2} h^{-1}) + 2, with r = 0.65 (excluding the March data from the sediments collected outside the mouth of the bay). While free oxygen is not directly used during denitrification, the correlation between oxygen consumption and denitrification rates likely reflects the close relationship between denitrification and nitrification rates. A large percentage of the benthic oxygen consumption is due to nitrification as discussed above. In addition, oxygen is used indirectly for nitrification since oxygen is consumed during the decomposition of organic matter which produces ammonium that is oxidized during nitrification.

The direct measurements of denitrification in Ochlockonee Bay during 1984–85 are considerably greater than the annual average denitrification rate estimated by Kaul & Froelich (1984) of 7.2 μg-at N m^{-2} h^{-1} based on a model of nitrate and salinity distributions in the water column in the bay during 1980–81. There are a number of possible reasons for the difference. It is not clear exactly what the difference would have been in the predicted denitrification rates if Kaul & Froelich's (1984) model had included ammonium or total nitrogen inputs and distributions. Also, river inputs of dissolved inorganic nitrogen during Kaul & Froelich's (1984) study were about 3 times lower than during a normal flow year, as discussed above. However, the rate of the internal input of nitrogen from benthic fluxes during 1980–81 was likely influenced by the mineralization of organic matter deposited during previous years when river inputs of nitrogen to the bay were greater. As such, there may have been a net internal benthic source of nitrogen to the bay during 1980–81 which would have masked the actual rate of denitrification. Direct measurements of denitrification and benthic NH_4^+ and NO_3^- + NO_2^- fluxes during a number of years when nitrogen input rates to the bay differ are needed to resolve this.

The riverine input of dissolved inorganic nitrogen (DIN) to Ochlockonee Bay (135 μg-at N m^{-2} h^{-1}) during years of normal river flow (in contrast to drought years when the input is ~45 μg-at N m^{-2} h^{-1}) is similar to the DIN input to a number of other estuaries that receive substantial anthropogenic nitrogen inputs, such as Narragansett Bay (108 μg-at N m^{-2} h^{-1}) and Delaware Bay (148 μg-at N m^{-2} h^{-1}), and is about twice as great as the DIN inputs to Chesapeake Bay (58 μg-at N m^{-2} h^{-1}) (DIN inputs as reported by Nixon & Pilson [1983]). The relatively high rate of nitrogen input to Ochlockonee Bay, in the absence of major anthropogenic nutrient inputs, is likely due to the large watershed area relative to the area of the bay compared to the above estuaries (Table 5). The ratio of watershed area to estuarine area is approximately 10 times greater for Ochlockonee Bay than many other estuaries, and results in a high freshwater input rate relative to the size of the estuary. The large freshwater input rate also leads to a short residence time for water in Ochlockonee Bay with the average during the study period being ca 3 d based on Kaul & Froelich's (1984) model of river flow *vs* residence time of water in Ochlockonee Bay. This contrasts with residence times on the order of weeks to months reported for many other estuaries.

Denitrification is a major sink for terrestrially derived nitrogen inputs to Ochlockonee Bay. The annual denitrification rate in Ochlockonee Bay was calculated by plotting the average denitrification rate in the bay as a function of month of collection and integrating the area under the curve. The magnitude of this sink for nitrogen was 73 μg-at N m^{-2} h^{-1}. A comparison of the annual riverine input of DIN during the study period (1 Apr 1984 to 31 Mar 1985) (135 μg-at N m^{-2} h^{-1}) with the annual denitrification rate measured during the same time interval (73 μg-at N m^{-2} h^{-1}) demonstrates that denitrification removed from the bay an amount of N equivalent to approximately 54 % of the riverine input of DIN (Table 6). This fairly high efficiency of removal of N via denitrification in an estuary with such

Table 5. Freshwater input rate, drainage basin area and estuarine surface area for a number of USA estuaries

Estuary	Freshwater input (m^3 s^{-1})	Drainage basin (10^3 km^2)	Estuary (km^2)	Area ratio Drainage : Estuary	Reference
Ochlockonee Bay	62.4	5.8	24	242	US Geological Survey unpubl. data*
Narragansett Bay, RI	105	4.7	328	14	Pilson 1985
Delaware Bay	447	33	1942	17	Sharp et al. 1982
Chesapeake Bay	2265	166	11300	15	Burger 1982, Wells et al. 1983
Neuse River Estuary, NC	55	11	400	28	Fisher et al. 1982
South River Estuary, NC	1	0.2	25	8	Fisher et al. 1982
Barataria Bay, LA		4	176	23	Day et al. 1982

* Area of estuary from planimetry of US Geological Survey maps No. 00624 and 01142

Table 6. Known sources and sinks of nitrogen in Ochlockonee Bay, Florida, April 1984 to April 1985

	Flux (μg-at N m^{-2} h^{-1})	Reference
Sources*		
Rivers (DIN)	135	This study
Precipitation	6	Hendry & Brezonik 1980
N_2-fixation	< 0.4	Seitzinger unpubl. data
Total	141	
Sinks		
Burial	?	
Export	?	
Denitrification	73	This study
Total	+73	

* Adequate data are not available to estimate offshore inputs. However, offshore inputs appear to be small relative to river inputs based on the following: annual average DIN in water at mouth of bay ~1 μM, flushing time of bay ~3 d, and volume of bay $\sim 2.4 \times 10^7$ m^3 Offshore input then calculated as (1 μg-at N l^{-1} $\times$ 2.4×10^{10} l $\times$ [0.0139 times h^{-1}])/2.4×10^7 m^2 = 14 μg-at N m^{-2} h^{-1}

a short water residence time (~3 d) may be related, in part, to the shallow water column (~1 m at low tide); such a shallow water column may increase the number of times N is cycled through the sediments and is subject to removal via denitrification, compared to deeper estuaries, before being transported offshore.

The importance of denitrification as a sink for nitrogen inputs to Ochlockonee Bay is similar to that in other estuaries for which data are available. In Narragansett Bay, denitrification removes an amount of nitrogen equivalent to 50 % of the DIN river and sewage inputs on an annual basis (Seitzinger et al. 1984). In the Tejo Estuary, Portugal, denitrification rates measured throughout the estuary during October (average 232 μg-at N m^{-2} h^{-1}; Seitzinger unpubl. data) were equivalent to approximately 45 % of the average DIN input from river-carried N and direct sewage sources to the estuary (516 μg-at N m^{-2} h^{-1}; Portuguese National Commission for the Environment unpubl. data). In Four League Bay (Louisiana, USA) denitrification removes an amount of N equivalent to 20 % of the river inputs of NO_3^- on an annual basis (Smith et al. 1985). In the Baltic Sea denitrification annually removes an amount of nitrogen equivalent to 40 % of the nitrogen inputs (Larsson et al. 1985). If the results from the above estuaries hold for estuaries in general, this implies that denitrification not only decreases the amount of N recycled for primary production within estuaries, but also effectively decreases, by as much as 40 to 50 %, the amount of river-supplied N (both natural and anthropogenic) transported to continental shelves and the open ocean.

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