

Direct denitrification in mangrove sediments in Terminos Lagoon, Mexico

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ABSTRACT: Rates of direct denitrification were measured using ¹⁵N isotope techniques in intact sediment cores from fringe and basin mangroves in Terminos Lagoon, Mexico. Sediments were injected with ¹⁵NO₃⁻ and the distribution of ¹⁵N was measured over time in the head space, overlying water, and sediments. Experiments included an investigation of spatial variation in denitrification rates by comparing results from the fringe and basin mangroves in the rainy season (July 1991); a second experiment was to determine the effect of 3 different NO₃⁻ concentrations (25, 100, 200 μmol core⁻¹ ¹⁵N-KNO₃⁻) on denitrification rates and was performed in the fringe mangrove during the 'Norte' season (January 1992). Highest ¹⁵N-N₂ fluxes were measured in the fringe mangrove at 9.4 μmol m⁻² h⁻¹, while denitrification rates in the basin mangrove ranged from 1.9 to 4.5 μmol m⁻² h⁻¹. ¹⁵N-N₂ fluxes in sediment cores from the fringe mangrove were significantly higher (4.5 to 7.7 μmol m⁻² h⁻¹) in cores enriched with 200 μmol core⁻¹ ¹⁵N-KNO₃⁻ compared to cores enriched with 25 and 100 μmol core⁻¹ ¹⁵N-KNO₃⁻ (<1 μmol m⁻² h⁻¹). Most of the applied ¹⁵N was recovered as particulate nitrogen in the sediment and a small fraction reduced to NH₄⁺ in both experiments. The low denitrification rates observed in the fringe and basin mangroves indicate that the capacity for sediment denitrification is limited by low NO₃⁻ availability. Previous nutrient exchange studies concluded that the fringe mangrove was a 'sink' of NO₃⁻ since sediment uptake of NO₃⁻ was assumed lost through denitrification. Results from this study show <10% of sediment NO₃⁻ uptake in fringe mangroves may be lost to denitrification; the remainder being immobilized in the sediment.

KEY WORDS: Mangroves · Denitrification · Nitrogen · Sediments · *Rhizophora* · *Avicennia* · Terminos Lagoon · Mexico

INTRODUCTION

Mangroves are a dominant feature of diverse coastal landscapes in the tropics (Por 1984) including river deltas, lagoons, estuaries, and carbonate platforms (Twilley et al. 1995). These forested wetlands are important to the productivity of tropical estuaries (Boto et al. 1985, Lugo et al. 1990, Twilley et al. 1992), yet

there is limited information about nutrient cycling in these intertidal macrophytes. Although nitrogen is an essential element to a variety of biological processes in mangrove forests (Alongi et al. 1992), there are few ecological studies of nitrogen transformations within mangrove ecosystems (Twilley 1988, Alongi et al. 1992). In particular, it is not clear how coupled nitrogen transformations within the forest influence the exchange of nitrogen at the boundary of mangroves with coastal waters.

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A few studies of nitrogen flux between mangroves and coastal waters conclude that mangroves may be a significant sink of dissolved inorganic nitrogen in tidal waters. For example, Walsh (1967) observed that NO_3^- concentrations entering a mangrove tidal creek in Hawaii, USA, decreased with distance inland, but no nitrogen fluxes were directly measured. Nedwell (1975) reported that about 90% of NO_3^- in a polluted station receiving sewage effluent was removed in a mangrove bordered tidal river in Fiji. Rivera-Monroy et al. (1995) measured nitrogen fluxes between Estero Pargo, an unpolluted tidal creek, and a fringe mangrove forest in Terminos Lagoon, Mexico, reporting that mangrove sediments were a sink of NO_3^- and NH_4^+ throughout the year. Denitrification, the dissimilatory reduction of NO_3^- to produce N_2O and N_2 , was considered the process that contributed to NO_3^- loss in these mangrove studies. Yet, there are few direct measurements of denitrification and estimates of how these rates influence the exchange of nitrogen in mangrove forests (e.g. Alongi et al. 1992). Denitrification may be an important process that regulates nitrogen flux at the mangrove-estuary boundary (Twilley 1988).

Denitrification is primarily dependent upon anoxic conditions, the presence of an energy source, and availability of NO_3^- substrate (Mosier & Schimel 1993). Depending on the NO_3^- source, there are 2 types of denitrification. Direct denitrification is fueled by NO_3^- that diffuses into sediments, while coupled denitrification is supported by NO_3^- produced by nitrification in sediments (Jenkins & Kemp 1984, Henriksen & Kemp 1988). Most of the denitrification studies in mangroves have focused on direct denitrification to understand the potential use of mangroves as a natural tertiary treatment of wastewater (Nedwell 1975, Corredor & Morell 1994). Corredor & Morell (1994) concluded that sediments in a fringe mangrove in Puerto Rico receiving secondarily treated sewage effluent were capable of denitrifying up to 15 times the normal NO_3^- concentrations (200 to 1000 μM). However, direct denitrification rates were low in sediments from an unpolluted mangrove tidal channel in Australia (Iizumi 1986) and represented minor losses of nitrogen from the ecosystem (Alongi et al. 1992). Boto & Wellington (1988) reported that NO_3^- was actually exported from this system, indicating a source of inorganic nitrogen to coastal waters.

These conflicting results on the denitrifying capacity of mangrove sediments lead to the confusion on the role of mangroves as a NO_3^- sink (Corredor & Morell 1994). There is practically no information on direct denitrification in mangroves under natural conditions (i.e. Iizumi 1986), nor how these rates may differ among riverine, fringe, or basin mangroves (Lugo & Snedaker 1974, Twilley 1988). In our study, we assumed that denitrification could account for the up-

take of NO_3^- observed in flux studies of mangroves by Rivera-Monroy et al. (1995), demonstrating the importance of this transformation to the fate of nitrogen in fringe and basin mangroves. We expected higher denitrification rates in the fringe than in the basin mangroves due to differences in NO_3^- availability. To test this hypothesis, we measured the effect of different NO_3^- concentrations on rates of denitrification in intact sediment cores from fringe and basin forests in Estero Pargo during 2 seasons using ^{15}N isotope techniques.

MATERIAL AND METHODS

Study area. This study was carried out in Terminos Lagoon ($18^\circ 40' \text{ N}$, $91^\circ 30' \text{ W}$), a large (~1800 km^2), shallow coastal lagoon located in the southwestern section of the Yucatan Peninsula in the state of Campeche, Mexico (Fig. 1). The climate of the area is tropical with annual average air temperatures ranging from 18 to 36°C . Tides are mixed diurnal with a mean tidal range of about 0.5 m. Average annual precipitation (1680 mm yr^{-1}) is seasonal, with a rainy season from June to October, which is associated with frequent tropical convective rains. The winter storm, or 'Norte' season, is from November to February with strong north winds and frontal rains. The dry season is from March to June. Peak river discharge occurs in the latter months of the rainy season from September to November.

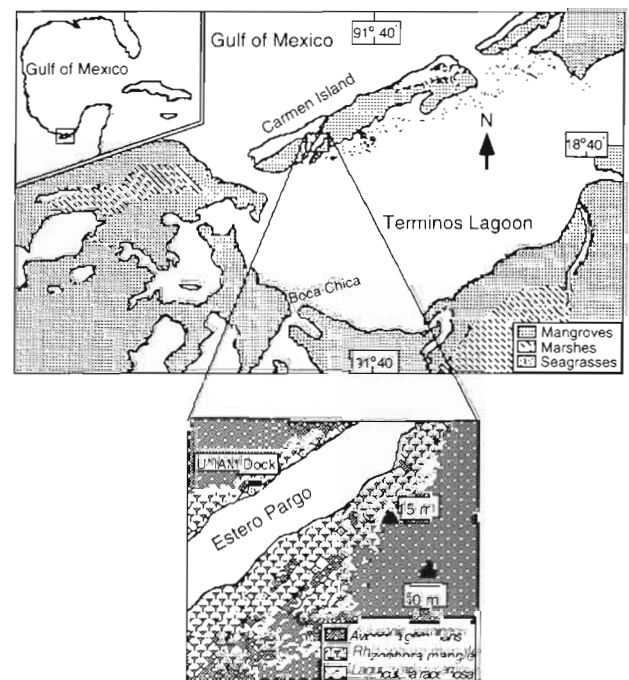


Fig. 1. Location map for sediment cores collected in the fringe and basin mangrove forests in Estero Pargo, Mexico.

The lagoon is bordered almost completely by extensive mangrove forests that are dominated by 3 species: *Rhizophora mangle* L. (red mangrove), *Avicennia germinans* L. (black mangrove), and *Laguncularia racemosa* Gaertn. f. (white mangrove) (Day et al. 1987). Estero Pargo is a tidal creek located on the lagoon side of the barrier island, Isla del Carmen. The forests adjacent to the tidal creek are characteristic of fringe mangroves with regular tidal inundation, while the inland forests are characteristic of basin mangroves which are infrequently flooded (Lugo & Snedaker 1974, Day et al. 1982). The bulk of the soil material in both mangroves consists of organic matter with many inclusions of fibrous mangrove roots and of coarser woody material. Bulk density in the fringe and riverine mangroves is 0.28 and 0.31 gdw cm⁻³, respectively (Lynch et al. 1989). Total nitrogen concentration in sediments reported for the fringe mangrove is 5.7 mg gdw⁻¹ and for the basin mangrove is 9 mg gdw⁻¹ (Lynch 1989). Terminos Lagoon and Estero Pargo have been described in detail elsewhere (Phlegher & Ayala-Castañares 1971, Ley-Lou 1985, Yañez-Arancibia & Day 1988).

Experimental design. To evaluate direct denitrification in mangrove sediments, 2 experiments were performed. The first experiment was to investigate spatial variation in denitrification rates by comparing results from the fringe and basin mangroves in the rainy season (July 1991). The second experiment was to determine the effect of different concentrations of NO₃⁻ on denitrification rates and was performed in the fringe mangrove during the 'Norte' season (January 1992). Similar concentrations of NO₃⁻ were used in the rainy and 'Norte' seasons in the fringe mangrove that allowed a temporal comparison of rates. Experiments were conducted in intact sediment cores collected about 15 m (fringe) and 50 m (basin) inland from the tidal creek in Estero Pargo (Fig. 1).

Four cores were collected from each of the 2 mangroves for the site comparison study, while 6 cores were used in the fringe mangrove for the concentration study. Plexiglas cylinders (30 cm long, 15 cm i.d. and 0.5 cm wall thickness) were carefully placed on the sediment surface and forced approximately 25 cm into the sediment. The cores were sampled in close proximity to each other to minimize spatial variability and were considered as replicates. Pneumatophores were present in all cores and precautions were taken to assure that the aerial portions of pneumatophores were not damaged during core sampling. Pneumatophore density was similar in both forests (428 ± 100 m⁻²) despite the dominance of *Rhizophora mangle* trees in the fringe mangrove. After collecting the core, a rubber cap was placed on the core base and secured with 2 aluminum bands. The cores were carefully transported in the dark back to the laboratory. Sediment tempera-

ture was measured during core sampling. Floodwater at each site was collected with previously acid-washed (10% HCl v/v) 1 l plastic containers.

Nitrogen-15 enrichment to the cores was 200 μmol core⁻¹ ¹⁵N-KNO₃ (99 atom% ¹⁵N) for the site comparison experiment in July 1991 (0.16 g N m⁻²). Cores collected in the fringe mangrove in January 1992 were randomly assigned to concentrations of either 25, 100, or 200 μmol core⁻¹ ¹⁵N-KNO₃ (2 cores per treatment). These enrichments represent <2% of the amount added in similar studies of wetland sediments (e.g. 19 g N m⁻², Reedy et al. 1989; 10 g N m⁻², Lindau & Delaune 1991). Concentrations used in our study were selected to assure that ¹⁵N emission reflected denitrification rates under natural conditions, yet provided a measurable ¹⁵N signal in the cores. Caffrey & Kemp (1992) used similar ¹⁵N enrichment of 0.15 g m⁻² to study direct denitrification in subtidal sediments. The isotope solutions were made with filtered (GF/F) floodwater collected in the field at the same time as the cores. Solutions were injected into each core at the surface, and at 2 and 9 cm depth to distribute the isotope throughout the core. A water depth of 1 to 2 cm was maintained over the sediment surface within the cores throughout the experiments with ambient floodwater. After isotope enrichment, the cores were incubated outdoors in the dark using a covered plastic chamber under ambient temperature (mean ambient temperature was 28°C).

In the site comparison experiment, 1 core was removed from the chamber at 1, 3, 5, and 8 d following ¹⁵N-enrichment. At that time pneumatophores were cut about 1 cm from the top of the core and covered with silicone grease (Corning®) to minimize altering the normal exchange of gases through these root structures (Scholander et al. 1955, Curran 1985, Nickerson & Thibodeau 1985, McKee et al. 1988). Production of N₂ was trapped in the headspace of each core by placing a plexiglas lid with gasket on the core top which was secured with 6 screws distributed evenly around the lid to form a gas-tight connection. The depth of each headspace was recorded, and zero-time gas samples were collected immediately after the headspace was sealed. Gas samples were taken from the headspace using a gas-tight syringe and hypodermic needle through a rubber septum sealed in the lid. Aliquots of 5 ml were withdrawn at 1 and 10 h, transferred to glass rubber-stoppered Vacutainers® (75 mm long by 10 mm i.d.), and stored until assay by ¹⁵N mass spectrometry. Vacutainers® were stored immersed in water to prevent contamination of atmospheric gases. After each headspace sampling, an equivalent volume (5 ml) of air was replaced to maintain pressure inside the core, and the core was returned to the incubation chamber. Changes in oxygen concentration in the

headspace throughout the 10 h incubation were determined on a second 5 ml aliquot sampled at the same time $^{15}\text{N}_2$ gas samples were taken. Oxygen and N_2 concentrations were measured with a gas chromatograph (SRI Instruments Model 8610) equipped with a TCD detector and a CTR dual-phase column (Alltech®) using helium as carrier gas. Oxygen concentrations changed <14% in all experiments and N_2 concentrations were constant over time (data not shown).

At the end of the 10 h time series, 50 ml of overlying water was sampled and each core sectioned at approximately 4 cm intervals to obtain 5 sediment samples per core. Replicate sediment subsamples (50 g per sample) were used for extraction of NH_4^+ and NO_3^- (Sumi & Koike 1990) with 2 N KCl (150 ml of KCl per 50 g of sediment) for 1 h. Another 2 subsamples (~35 g) were also taken from each sample for analysis of percent water (dried at 60°C for 48 h) and bulk density. Dry sediments were ground with mortar and pestle, and assayed for total carbon and nitrogen and ^{15}N content in 4 subsamples per sample.

Sampling of headspace and sediment during the NO_3^- concentration study followed a similar procedure as the site comparison study. Headspace samples were collected at 1 and 10 h periods following ^{15}N application and after sealing the headspace in each core. Headspace sampling was repeated in each core at 1, 3, and 5 d. At the end of the 5 d incubation the 6 cores were then sectioned and processed for nutrient and ^{15}N analyses as described above.

Sample analysis. Total carbon and particulate nitrogen (PN) were analyzed with a LECO® CHN elemental analyzer. Overlying water and KCl extracts were analyzed for NH_4^+ (Solórzano 1969) and NO_3^- (Grasshoff et al. 1983) concentrations. Nitrogen concentrations and sediment densities were used to calculate total NO_3^- , NH_4^+ , and PN per core. Headspace samples were assayed for nitrogen isotopic masses of 28, 29, and 30 by injecting directly into a nuclide Model 3-60-RMS double collector instrument (Premier American Technologies Corp., Bellefonte, PA, USA). Initial air samples obtained at 0 h during the 10 h time series sampling were analyzed as a reference gas (background) on the IRMS inlet using the sample inlet (Mulvaney & Kurtz 1982).

^{15}N enrichment in overlying water, KCl extracts and sediment were measured with a JASCO ^{15}N emission spectrometer. $^{15}\text{NH}_4^+$ and $^{15}\text{NO}_3^-$ in the overlying water and KCl extracts were isolated by sequential steam distillations using MgO to raise the pH above 9.0 (Keeney & Nelson 1982) following reduction of $\text{NO}_3^- + \text{NO}_2^-$ to NH_4^+ by Devarda's alloy (Bremer 1965). Carry-over between samples was minimized by a distillation of ethanol between each sample (Fiedler & Proksch 1975). Fifty ml of the condensate was collected directly

onto approximately 0.035 g of ion sieve (Union Carbide #W-85). The ion sieve trapped 95% of the NH_4^+ based on preliminary experiments with 50 ml of distillate. The ion sieve was collected on a 25 mm A/E filter, dried (60°C), placed in Petri dishes, and stored in a vacuum desiccator until assay for ^{15}N content. Recovery of NH_4^+ and NO_3^- following this distillation procedure averaged 99% and 75%, respectively. Samples were prepared by first converting organic and inorganic nitrogen to nitrogen gas with a dry microDumas combustion technique (Fielder & Proksch 1975). Sediment and ion sieve samples were placed in previously degassed (550°C, 12h) discharge tubes (Pyrex glass 6.0 mm O.D.) containing CaO and Cuprox. The tubes were evacuated to 1 mTorr, and filled with argon to 1 Torr, and preheated to remove residual water vapor. Tubes were again evacuated to 1 mTorr and argon added to obtain vacuum of 1 Torr. The tubes were sealed and later combusted at 550°C for 12 h and cooled to room temperature for another 12 h. The atom% ^{15}N for samples was then determined with a JASCO ^{15}N analyzer (Model N-150). On each sample, 3 scans of the light emission spectra of the 28 and 29 mass were performed. The instrument was calibrated with commercial standards prepared from samples analyzed by mass spectrometry (Japan Spectroscopic Co., Instruction Manual model N, 1986). The formula used to calculate atom% was:

$$^{15}\text{N}(\text{atom}\%) = \frac{100}{2R+1} \text{ where } R = \frac{^{14}\text{N}_2}{^{15}\text{N}}$$

(Fielder & Proksch 1975).

Denitrification rates were calculated using a mass spectrometric procedure developed for determination of ^{15}N - N_2 evolved from sediments treated with ^{15}N -labeled substrates (Mulvaney & Kurtz 1982, Mulvaney & Kurtz 1984, Mulvaney & Boast 1986). The equations used to calculate the total N-flux from the sediment in each core were:

$$\Delta r = \left(\frac{^{29}\text{N}_2}{^{28}\text{N}_2} \right)_{\text{sample}} - \left(\frac{^{29}\text{N}_2}{^{28}\text{N}_2} \right)_{\text{reference}}$$

$$\Delta r' = \left(\frac{^{30}\text{N}_2}{^{28}\text{N}_2} \right)_{\text{sample}} - \left(\frac{^{30}\text{N}_2}{^{28}\text{N}_2} \right)_{\text{reference}}$$

$$^{15}\text{X}_\text{N} = \frac{2 \Delta r'}{\Delta r + 2 \frac{\Delta r'}{\Delta r}}$$

$$d = \frac{\Delta r'}{\{^{15}\text{X}_\text{N}\}^2}$$

$$\text{N}_2 \text{ flux} = \frac{\Delta C}{A \times \Delta t}$$

where 29/28 and 30/28 are ion current ratios determined by the mass spectrometer; 'sample' is air sample

from a core at some time, t , after installation of the core lid; 'reference' is a normal air sample taken from the core immediately after core lid installation; $^{15}X_N$ = mole fraction of ^{15}N in the soil NO_3^- pool; d is the fraction of total N gas in the core headspace attributable to denitrification; A is the area of the core; Δt is the time the lid covered the core; and ΔC is the change in the amount of $^{30}N_2$ and $^{29}N_2$ in the core headspace during time t [$\Delta C = (\text{total } N_2 \text{ in the core headspace} \times d)_{t_1} - (\text{total } N_2 \text{ in the core headspace} \times d)_{t_0}$] (Mosier & Schimel 1993).

Significant differences in rates of $^{15}N-N_2$ production among treatments in the NO_3^- concentration experiment were determined with a 1-way ANOVA with a level of significance of 0.05 (SAS 1993). A 2-way analysis of variance was used to test differences in sediment NH_4^+ and NO_3^- concentrations between areas in the site comparison experiment.

RESULTS AND DISCUSSION

Nutrient concentrations and atom% ^{15}N enrichment

Mean NH_4^+ concentrations were significantly higher ($p < 0.05$) in the fringe than in the basin mangroves in July 1991 (Table 1, Fig. 2). Concentrations ranged from

Table 1. Bulk density (BD $g\ cm^{-3}$), carbon to nitrogen ratios (C:N), and extractable-N ($\mu g\ gdw^{-1}$) averaged with depth (0 to 23 cm) in cores from fringe and basin forests incubated at different days in July 1991; mean values ($\pm 1\ SD$)

Forest type	Parameter	Incubation time (d)			
		1	3	5	8
Fringe	BD	0.20 (0.01)	0.26 (0.01)	0.20 (0.01)	0.20 (0.01)
	C:N	25.1 (5.1)	22.1 (1.7)	19.0 (0.61)	21.3 (1.5)
	NH_4^+	35.3 (12.2)	28.0 (15.5)	37.2 (8.11)	37.1 (14.5)
	NO_3^-	6.7 (1.4)	4.5 (2.9)	7.0 (3.01)	11.4 (6.0)
Basin	BD	0.22 (0.01)	0.4 (0.01)	0.22 (0.01)	0.60 (0.02)
	C:N	22.3 (1.2)	23.6 (1.7)	22.8 (1.2)	39.2 (16.0)
	NH_4^+	14.7 (5.9)	11.9 (10.0)	8.3 (6.8)	9.1 (1.9)
	NO_3^-	3.7 (0.8)	4.1 (0.9)	2.4 (1.7)	3.2 (1.5)

22 to 44 $\mu g\ gdw^{-1}$ in the fringe mangrove and from 6 to 18 $\mu g\ gdw^{-1}$ in the basin mangrove. There was a decline from 18 $\mu g\ gdw^{-1}$ at 0 to 6 cm to 7 $\mu g\ gdw^{-1}$ at 18 to 23 cm in the basin mangrove (Fig. 2). NH_4^+ concentrations did not show any pattern with depth in the fringe mangrove. Although NO_3^- concentrations were higher in the fringe than in the basin mangrove, this difference was not significant ($p > 0.05$). Mean NO_3^- concentrations ranged from 5.5 to 9.0 $\mu g\ gdw^{-1}$ in the fringe mangrove and from 3.5 to 6.5 $\mu g\ gdw^{-1}$ in the basin mangrove. There were no differences in NO_3^- concentrations with depth in either area. Particulate nitrogen (PN) concentrations decline with depth in both mangroves (Fig. 3). PN decreased from 11 $mg\ gdw^{-1}$ at 0 to 5 cm to 5.5 $mg\ gdw^{-1}$ at 17 to 21 cm in the

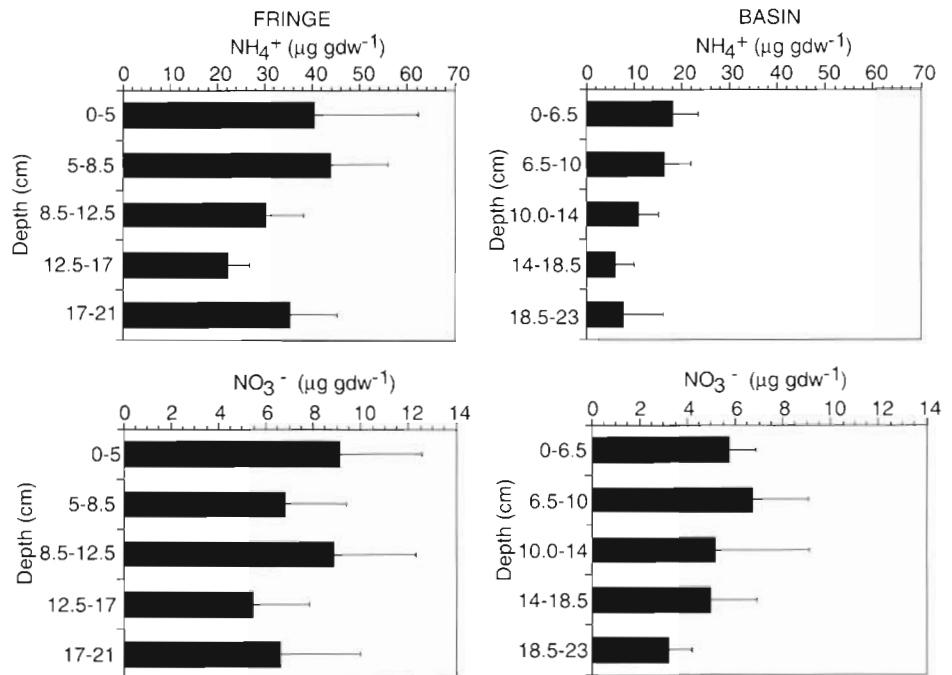


Fig. 2. NH_4^+ and NO_3^- concentrations with depth in cores from the fringe and basin forest in July 1991

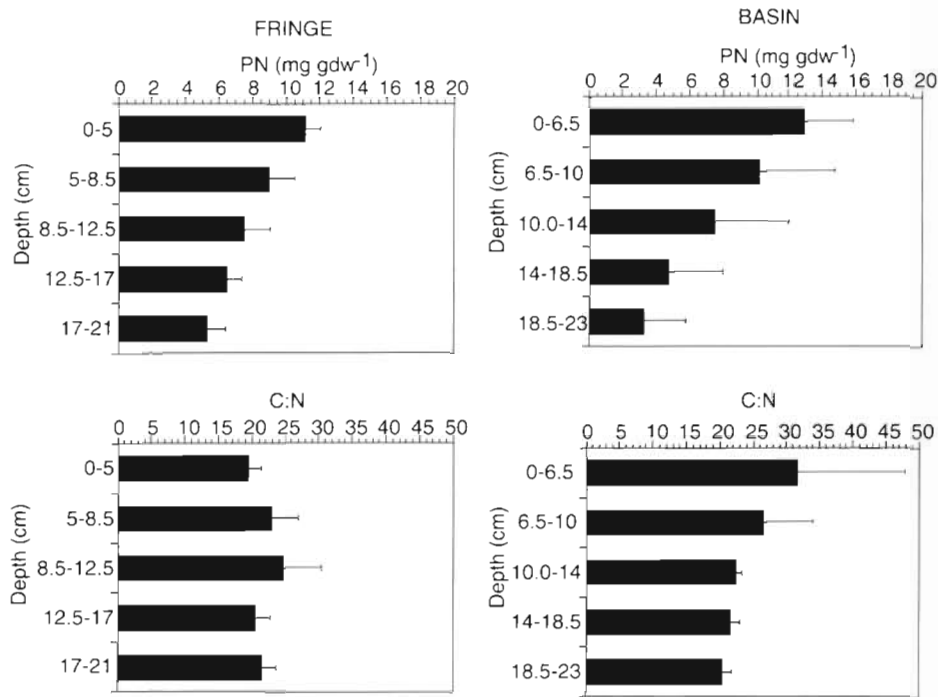


Fig. 3. Particulate nitrogen (PN) and carbon to nitrogen ratios (C:N) with depth in cores from the fringe and basin forest in July 1991

fringe mangrove. In the basin mangrove there was a large variation associated with the mean at each depth, and concentrations declined from 12.5 mg gdw⁻¹ to 3 mg gdw⁻¹. PN concentrations were higher in the top 10 cm in the basin mangrove than in the fringe mangrove. C:N ratios were not significantly different with depth in either mangrove, although a slightly higher ratio was measured in the basin mangrove in the top 10 cm. Ratios ranged from 19 to 25 in the fringe mangrove and from 21 to 32 in the basin mangrove.

Mean NH₄⁺ concentrations were significantly lower ($p < 0.05$) in duplicate cores treated with 25 $\mu\text{mol core}^{-1}$ ¹⁵N-KNO₃ than in cores treated with 100 and 200 $\mu\text{mol core}^{-1}$ ¹⁵N-KNO₃. Mean NO₃⁻ concentrations were higher in cores amended with 200 $\mu\text{mol core}^{-1}$ ¹⁵N-

KNO₃, but this difference was not significant (Table 2). There were no differences in mean C:N ratios and bulk densities in all treatments (Table 2). Mean NO₃⁻ concentrations with depth were higher in cores enriched with 200 $\mu\text{mol core}^{-1}$ ¹⁵N-KNO₃ with values of 7.5 to 9 $\mu\text{g gdw}^{-1}$ at 12 to 19 cm, compared to 2 or 3 $\mu\text{g gdw}^{-1}$ for the other 2 treatments (Fig. 4). Mean atom% ¹⁵NO₃⁻ and ¹⁵NH₄⁺ enrichment with depth for all treatments ranged from 0.1 to 0.31 and 0.1 to 0.39, respectively (Table 2).

¹⁵N₂ production

Denitrification rates were extremely low in both the fringe and basin mangroves in July 1991, although sediments from both areas were treated with 200 $\mu\text{mol core}^{-1}$ ¹⁵N-KNO₃ (Fig. 5). The highest denitrification rate (9.4 $\mu\text{mol m}^{-2} \text{h}^{-1}$) was observed in the fringe mangrove after 3 d of incubation. This was the only flux of labeled N₂ observed in this mangrove during the 8 d incubation period. In contrast, ¹⁵N₂ fluxes in the basin mangrove declined from 4.5 $\mu\text{mol m}^{-2} \text{h}^{-1}$ after Day 1 to 1.9 $\mu\text{mol m}^{-2} \text{h}^{-1}$ at Day 8. No fluxes were observed at this site on Days 3 and 5. Rates <5 $\mu\text{mol m}^{-2} \text{h}^{-1}$ are within the lower limits of detection given the analytical sensitivity and size of headspace in these experiments. Thus the presence and absence of rates in Fig 5 indicate the generally low rates of denitrification at the limits of detection at both mangrove sites.

Table 2. Bulk density (BD g cm⁻³), carbon to nitrogen ratios (C:N), extractable-N ($\mu\text{g gdw}^{-1}$) averaged with depth (0 to 20 cm), and atom% excess in duplicated cores from a fringe forest enriched with different ¹⁵NO₃⁻ concentrations ($\mu\text{mol core}^{-1}$) in January 1992; mean values (± 1 SD)

Parameter	¹⁵ NO ₃ ⁻ treatment concentration		
	25	100	200
BD	0.22 (0.01)	0.26 (0.06)	0.21 (0.01)
C:N	16.8 (0.6)	18.6 (2.0)	17.4 (0.2)
NH ₄ ⁺	12.9 (7.8)	24.5 (4.0)	22.9 (8.7)
NO ₃ ⁻	2.7 (1.6)	2.9 (1.8)	6.2 (3.2)
atom% ¹⁵ NO ₃ ⁻	0.1 (0.2)	0.3 (0.1)	0.2 (0.1)
atom% ¹⁵ NH ₄ ⁺	0.1 (0.1)	0.4 (0.4)	0.2 (0.2)

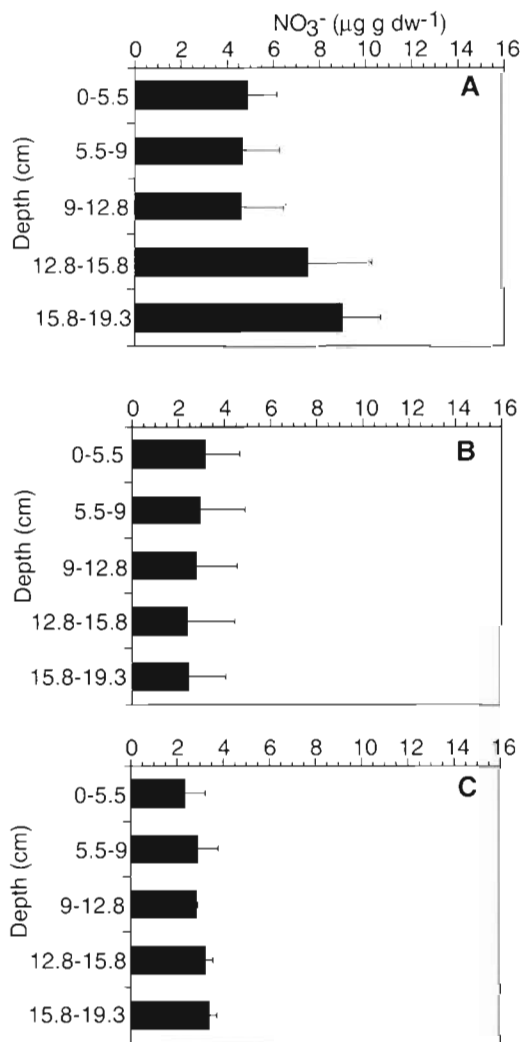


Fig. 4. NO_3^- concentrations with depth in cores from the fringe forest enriched with different $^{15}\text{N-KNO}_3$ concentrations in January 1992 as follows: (A) $200 \mu\text{mol core}^{-1}$, (B) $100 \mu\text{mol core}^{-1}$, (C) $25 \mu\text{mol core}^{-1}$

Spatial and temporal variability were confounded in our study since we only analyzed 1 core per time during the incubation in July 1991. Similar concentrations of extractable nitrogen were observed in all cores from the basin mangrove (Table 1), yet the different physical properties of sediments in these experiments may have influenced the timing of $^{15}\text{N}_2$ fluxes. However, despite these problems, it is apparent that denitrification rates are low in the basin forest. On the other hand, similar rates in replicate cores from the enrichment experiment in January 1992 indicate that most of these experimental cores represented homogenous sediment conditions. Results from this experiment also indicate that nitrogen loss through denitrification may be low in the fringe forest. Nielsen et al. (1990) pointed

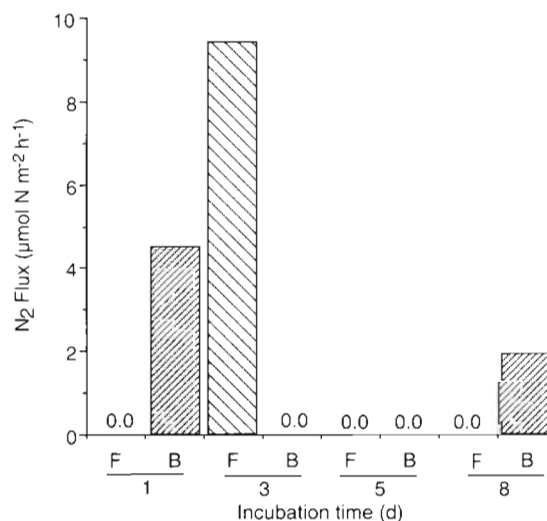


Fig. 5. Denitrification rates at different incubation times in sediment cores from the fringe (F) and basin (B) mangrove forests in Estero Pargo, Mexico in July 1991

out that even when there are high densities of denitrifiers in sediment, diffusion processes influenced by bulk density (Rosenfeld 1979, Mackin & Aller 1984) can control the supply of electron donors or acceptors, even when such nutrients are present at high concentrations. Lindau & Delaune (1991) and Risgaard-Petersen et al. (1994) found high variation in N_2 fluxes among replicates and attributed it to the nonhomogeneity of cores used in their experiments. Kessel et al. (1993) stated that the major problem in estimating N losses by denitrification was the high degree of spatial and temporal variability of this process.

Gas 'entrapment' may contribute from 16 to 41% of the applied ^{15}N to the mass balance of nitrogen in permanently flooded sediment systems (Lindau et al. 1988, Katyal et al. 1989, Lindau & Delaune 1991). These studies found significant reservoirs of $^{15}\text{N}_2$ in the porewaters from 16 to 33 d following amendments of either urea or $^{15}\text{NO}_3^-$ to the sediment. Estimates using Bunsen solubility coefficients (Weiss 1970) indicate that N_2 concentrations are negligible in cores where $^{15}\text{N}_2$ in the headspace was detected. It is not possible to determine if gas 'entrapment' was an important process if the nitrogen flux to the headspace was not in equilibrium with the porewater. This difference between the 2 pools is difficult to evaluate due to the lack of direct $^{15}\text{N}_2$ gas measurements in the pore and overlying water. However, even assuming that $^{15}\text{N}_2$ concentrations in the pore and overlying water are not in equilibrium, the high ^{15}N concentrations found in the total sediment pool at the end of the incubation period (see below) do indicate that ^{15}N gas production trapped in the sediment might have been low.

Pneumatophores inside our cores were cut and sealed with silicone grease to avoid altering exchange of gases and oxidation in the sediment root zone during the 8 h incubations. Pneumatophores have a significant amount of aerenchyma tissue and are considered as organs responsible for gas exchange between the atmosphere and the internal tissue of mangroves (Scholander et al. 1955). Redox measurements in areas vegetated by this species indicate that this supply of O₂ can modify the redox status of normally reduced sediments (Thibodeau & Nickerson 1986, Carlson & Yabro 1988, McKee et al. 1988, McKee 1993). Recent redox measurements in cores from a fringe mangrove in Florida, incubated as described in this study, and with pneumatophore densities similar to the ones observed in our cores, show that redox potentials do not significantly change during 5 d of incubation (Rivera-Monroy et al. unpubl. results).

Mass balance

The contribution of N₂ production to the mass balance of enriched nitrogen for each core in both experiments was calculated from the cumulative production of N₂ during the entire incubation period based on hourly denitrification rates. The recovery of ¹⁵N in the site comparison experiment between fringe and basin mangroves was <55% after 8 d of incubation (Table 3). These estimates of ¹⁵N recovery do not include ¹⁵N in larger roots and pneumatophores, since these nitrogen pools were not measured. The largest ¹⁵N fraction was measured in the sediments with recovery ranging from 33 to 47% in the fringe mangrove, and from 38 to 50% in the basin mangrove. The second largest ¹⁵N fraction was measured in the extractable NH₄⁺ pool. ¹⁵NH₄⁺ represented <3% of the total ¹⁵N recovered in the

fringe mangrove and <5% in the basin mangrove. ¹⁵NO₃⁻ concentrations were <1% in both mangroves (Table 3). Similarly, the amount of ¹⁵NO₃⁻ and ¹⁵NH₄⁺ in the overlying water was almost zero and represented a very small fraction of the total ¹⁵N added in cores. Caffrey & Kemp (1992) also found a small fraction (<1%) of ¹⁵N in the overlying water of vegetated cores. ¹⁵N₂ in the headspace was detected after 3 d in the fringe mangrove (6 μmol) and after 1 d in the basin mangrove (2 μmol). This flux of nitrogen represented <3% of the recovered ¹⁵N.

Mass balance in the fringe and basin mangroves suggests that there are other ¹⁵NO₃⁻ sinks. Since we used intact cores, it is possible that some ¹⁵NO₃⁻ was absorbed by the root system. Christensen et al. (1990) pointed out that the presence of plants and their roots can make NO₃⁻ unavailable to denitrifying bacteria through uptake and assimilation. Boto et al. (1985) reported that in laboratory experiments NO₃⁻ uptake was critical for fine root development of *Avicennia marina* seedlings. Also, NO₃⁻ uptake by algae attached to pneumatophores (Dor & Levy 1984, Rodriguez & Stoner 1990) could be another NO₃⁻ sink inside the cores. Apparently, 'competition' for NO₃⁻ between denitrifiers and uptake by these nitrogen pools might be important in the fringe and basin mangroves during the rainy season.

As in the site comparison study, sediments in the substrate concentration study had the largest accumulation of ¹⁵N (Table 4). The recoveries of excess ¹⁵N for both cores enriched with 200 μmol core⁻¹ ¹⁵N-KNO₃ were 70 and 98%, higher than values in the site comparison study. For the replicated cores enriched with 100 μmol core⁻¹ ¹⁵N-KNO₃ the recoveries were 99 and 104%, and for the cores in the 25 μmol core⁻¹ ¹⁵N-KNO₃ treatment the recoveries were lower at 41 and 80% (Table 4). The second largest fraction of ¹⁵N

Table 3. Distribution of applied ¹⁵N (μmol core⁻¹) among various forms of nitrogen including nitrogen gas (N₂), extractable N (NH₄⁺ and NO₃⁻), sediment nitrogen and total ¹⁵N recovered at different times following enrichment as an average with depth (~0 to 20 cm) in cores from fringe and basin mangroves in the rainy season, July 1991. Mean (±1 SD). -: no data

Date	Forest type	Time (d)	Applied N (μmol core ⁻¹)	Total N ₂ ^d headspace	Extractable N ^{tb}		Overlying water		Sediment ^a	Total
					NH ₄ ⁺	NO ₃ ⁻	NH ₄ ⁺	NO ₃ ⁻		
4 July 1991	Fringe	1	200	0	3 (0.5)	0.06 (0.03)	-	-	67 (17)	69.3
		3	200	6	4 (1.3)	0.13 (0.02)	0	0	68 (23)	78.1
		5	200	8	6 (0.6)	0.07 (0.05)	-	-	78 (29)	92.1
		8	200	8	5 (1.5)	0.07 (0.02)	0	0	94 (35)	107.4
7 July 1991	Basin	1	200	2	9 (2.7)	0.07 (0.03)	0	0	77 (14)	88.1
		3	200	3	5 (1.3)	0.02 (0.01)	0	0	97 (43)	105.0
		5	200	3	7 (37)	0.20 (0.05)	-	-	100 (34)	110.2
		8	200	5	3 (0.7)	0.01 (0.00)	-	-	95 (16)	103.1

^a Calculated by interpolating denitrification rates (see 'Methods')

^b Includes all 5 sections per core.

Table 4. Distribution of applied ^{15}N ($\mu\text{mol core}^{-1}$) among various forms of nitrogen including nitrogen gas (N_2), extractable N (NH_4^+ and NO_3^-), sediment nitrogen and total ^{15}N recovered in duplicate cores after a 5 d incubation following enrichment as an average with depth (~0 to 20 cm) in cores from the fringe forest in the 'Norte' season, January 1992. Mean (± 1 SD). -: no data

Applied N ($\mu\text{mol core}^{-1}$)	Core ID	Total N_2^{a} headspace	Extractable N^{b}		Overlying water		Sediment ^c	Total
			NH_4^+	NO_3^-	NH_4^+	NO_3^-		
200	1	3.7	0.1 (0.0)	0.5 (0.0)	-	-	135.7 (57.4)	140.1
200	2	6.5	1.5 (0.0)	0.4 (0.0)	-	-	187.9 (45.5)	196.4
100	1	0.0	13.8 (1.4)	3.5 (0.5)	-	-	86.7 (42.0)	104.0
100	2	0.5	1.1 (0.1)	2.1 (0.3)	-	-	95.0 (40.0)	98.7
25	1	0.0	0.1 (0.0)	0.0 (0.0)	-	-	10.3 (14.6)	10.4
25	2	0.0	0.6 (0.0)	0.0 (0.0)	-	-	19.1 (13.0)	19.7

^aCalculated by interpolating denitrification rates (see 'Methods')

^bIncludes all 5 sections per core

recovered in this experiment was the exchangeable nitrogen pool. The highest recovery (14%) of extractable $^{15}\text{NH}_4^+$ was measured in one of the cores enriched with $100 \mu\text{mol core}^{-1}$ $^{15}\text{N-KNO}_3$, but overall the recovery of ^{15}N in this pool was <10%. $^{15}\text{NO}_3^-$ recoveries in the KCl-extracts for all cores were <4%. $^{15}\text{N}_2$ in the headspace represented <3% of the recovered ^{15}N . $^{15}\text{NH}_4^+$ and $^{15}\text{NO}_3^-$ in the overlying water were not determined in this experiment due to analytical problems. The recovery of extractable nitrogen pool in the $100 \mu\text{mol core}^{-1}$ $^{15}\text{N-KNO}_3$ and $200 \mu\text{mol core}^{-1}$ $^{15}\text{N-KNO}_3$ enrichments may be underestimated due to missing values at some depths. However, this underestimate does not change the overall amount of recovery since low ^{15}N concentrations were generally found in the extractable nitrogen pool.

Recovery of ^{15}N in the NO_3^- enrichment experiment in January 1992 was >80%, higher than the site comparison study in July 1991. In both experiments most of the recovered ^{15}N in all cores was measured in the sediment. The low recovery of ^{15}N in the extractable pool (which includes pore water ^{15}N and exchangeable ^{15}N ; Mackin & Aller 1984) indicates that adsorption of ^{15}N was low. This suggests that a large amount of inorganic nitrogen was probably absorbed by sedimentary bacteria. Davidson et al. (1992) concluded that microbial assimilation of NO_3^- may be an important pathway for NO_3^- retention in forest ecosystems. This might be the case in Estero Pargo given the low NO_3^- concentrations observed in both mangroves (Table 1). When the C:N ratio of substrate is low (<15:1) decomposers are not N limited (Morris 1991), and a net release of inorganic nitrogen to the sediment solution occurs (i.e. mineralization). The high C:N ratios measured in sediments in the fringe (19 to 25.1) and basin (22.3 to 39.2) mangroves in Estero Pargo (Table 1) indicate that nitrogen assimilation by bacterial populations in the sediments was probably favored over mineralization.

We used a mass balance approach to estimate potential denitrification rates in comparison to rates calculated from $^{15}\text{N-N}_2$ production in the cores amended with $200 \mu\text{mol core}^{-1}$ $^{15}\text{N-KNO}_3$. Assuming that the unrecovered ^{15}N after 8 d of incubation is lost to denitrification in the fringe ($92.6 \mu\text{mol}$) and basin ($96.9 \mu\text{mol}$) sediments in July 1991, then we would have denitrification rates of $26.5 \mu\text{mol m}^{-2} \text{h}^{-1}$ and $27.8 \mu\text{mol m}^{-2} \text{h}^{-1}$, respectively. Similarly, for replicate cores amended with $200 \mu\text{mol}$ in January 1992, we calculated denitrification rates of 27.5 and $1.6 \mu\text{mol m}^{-2} \text{h}^{-1}$. It is likely that most of the unaccounted ^{15}N may have accumulated in other nitrogen pools (i.e. pneumatophores with attached algae) and not lost to denitrification. The high recovery (70 to 100%) of ^{15}N in the sediments and the low denitrification rates also indicate that sedimentary microbes may play an important role in the nitrogen cycle of the fringe and basin mangroves. These results are similar to other studies that demonstrate the significance of microbial immobilization of NO_3^- in forest soils (Davidson et al. 1992, Hart et al. 1994).

We observed $^{15}\text{NH}_4^+$ in KCl extracts (Tables 3 & 4) from sediment in both experiments indicating that NO_3^- 'ammonification', the reduction of NO_3^- to NH_4^+ (Koike & Sørensen 1988), occurred in all the incubations. In contrast to denitrification, which represents a net loss of nitrogen from an ecosystem, NO_3^- 'ammonification' recycles nitrogen within the ecosystem as NH_4^+ . In general, denitrifiers dominate environments in which the C:N ratio is low (<15), whereas NO_3^- is primarily reduced to NH_4^+ in carbon rich environments (C:N > 20) (Cole & Brown 1980). Denitrification and NO_3^- ammonification can occur simultaneously in sediments; the dominance of one process over the other depends on NO_3^- concentrations and C:N ratios of the sediments (Smith et al. 1982, King & Nedwell 1987, Rehr & Klemme 1989). NO_3^- ammonification may be an important nitrogen transformation in mangrove

ecosystems due to their generally low NO_3^- sediment concentrations (pore waters = 0 to 21 μM ; Alongi et al. 1992, Boto 1992) and high C:N ratios (>20) (Boto & Wellington 1984, Twilley et al. 1986, Steyer 1988, Lynch et al. 1989, Lugo 1990). Certainly, NO_3^- reduction to NH_4^+ in temperate coastal sediments is equally or sometimes more important than denitrification (Sørensen 1978, Nishio et al. 1982, Koike & Sorensen 1988). Despite the potential key role NO_3^- ammonification might have in the nitrogen cycle in mangrove ecosystems, there are no published references on this process.

Denitrification and NO_3^- exchange

$^{15}\text{N}_2$ fluxes from sediment cores in the fringe mangrove treated with different concentrations of NO_3^- were distinct (Fig. 6). After 1 d of incubation, denitrification rates ranged from 4.5 to 7.7 $\mu\text{mol m}^{-2} \text{h}^{-1}$ in the 200 $\mu\text{mol core}^{-1}$ $^{15}\text{N-KNO}_3$ treatment, but $^{15}\text{N}_2$ production was not observed at 25 and 100 $\mu\text{mol core}^{-1}$ $^{15}\text{N-KNO}_3$ during the initial 24 h of incubation. Rates declined by approximately 80% in both cores enriched with 200 $\mu\text{mol core}^{-1}$ $^{15}\text{N-KNO}_3$ after 3 d of incubation. The only observed production of $^{15}\text{N}_2$ in the other 2 treatments was measured in one core at 100 $\mu\text{mol core}^{-1}$ $^{15}\text{N-KNO}_3$, while no N_2 flux was observed during the 5 d experiment in either of the 2 cores amended with 25 $\mu\text{mol core}^{-1}$ $^{15}\text{N-KNO}_3$. Thus, rates were significantly higher ($p < 0.05$) in cores enriched with 200 $\mu\text{mol core}^{-1}$ $^{15}\text{N-KNO}_3$, while there was no significant

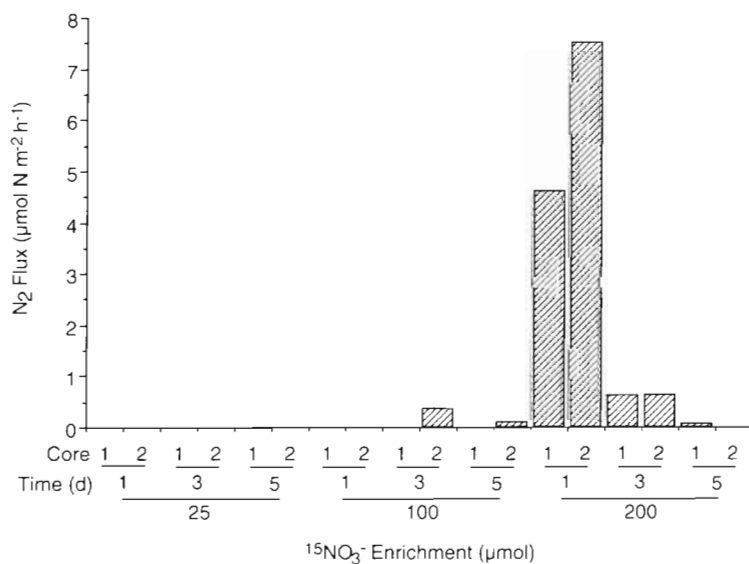


Fig. 6. Denitrification rates at different NO_3^- enrichments and incubation time in sediment cores collected in the fringe forest in January 1992 in Estero Pargo, Mexico

difference in the 25 and 100 $\mu\text{mol core}^{-1}$ $^{15}\text{N-KNO}_3$ amendments. However, by Day 5 denitrification rates were 0.08 $\mu\text{mol m}^{-2} \text{h}^{-1}$ in one core at 200 $\mu\text{mol core}^{-1}$, and 0.11 $\mu\text{mol m}^{-2} \text{h}^{-1}$ in the 100 $\mu\text{mol core}^{-1}$ enrichment.

The spatial and seasonal variation of denitrification in marine sediments has been associated with both the kinetic nature of denitrification capacity and differences in NO_3^- concentration (Twilley & Kemp 1986). The kinetic response of denitrification in mangrove sediments to NO_3^- concentration has been investigated indirectly using NO_3^- uptake (reduction) from overlying water (Nedwell 1975, Iizumi 1986, Shaiful 1987). The kinetic nature of NO_3^- uptake (potential denitrification) is described with K_m values that represent NO_3^- concentrations at one-half the saturated uptake (reduction) rates. K_m values (μM) for mangrove sediments vary from a low of 10 to 69 μM for mangroves in Hinchinbrook Island, Australia (Iizumi 1986), to higher values of 167 to 189 μM for mangroves in Selangor, Malaysia (Shaiful 1987), and 180 to 600 μM in mangroves impacted by sewage enrichment in Fiji (Nedwell 1975). Mean denitrification rate at Hinchinbrook Island was 0.53 $\mu\text{mol m}^{-2} \text{h}^{-1}$ (Iizumi 1986) compared to a range of 78 to 261 $\mu\text{mol m}^{-2} \text{h}^{-1}$ in the polluted site in Fiji (Nedwell 1975). Maximum NO_3^- reduction rates (V_{max} using a kinetic approach) in mangrove sediments using a flow through core design were 1320 to 1520 $\mu\text{mol m}^{-2} \text{h}^{-1}$ in 2 mangrove sites in Selangor (Shaiful 1987). The extreme variation in the kinetic nature of potential denitrification (NO_3^- uptake) in mangrove sediments follows the pattern of lower affinity (higher K_m) and higher capacity (higher V_{max}) in areas of higher carbon and nitrogen supply (Twilley & Kemp 1986).

The highest rates of NO_3^- uptake by mangrove sediments in Estero Pargo were about 179 $\mu\text{mol m}^{-2} \text{h}^{-1}$ (Rivera-Monroy et al. 1995), similar to the medium range of rates measured in mangrove sediments described above by Nedwell (1975). Maximum rates of NO_3^- uptake in mangrove sediments, using the flume technique, occurred in June 1991 at the beginning of the rainy season and were associated with elevated NO_3^- concentrations in the river (Rivera-Monroy et al. 1995). Rivera-Monroy et al. (1995) assumed that NO_3^- reduction in waters flooding the fringe mangrove was lost through denitrification, and concluded that this fringe mangrove was a sink of nitrogen. However, as shown by our ^{15}N experiments, denitrification accounted for $<10\%$ of the applied $^{15}\text{NO}_3^-$, suggesting that this mangrove acts as a

transformer of nitrogen (Nixon & Lee 1986) rather than a nitrogen sink.

This is the first work where ^{15}N techniques have been used to evaluate denitrification rates in mangrove sediments. Addition of inorganic $^{15}\text{NO}_3^-$ to sediment cores from fringe and basin mangroves in Estero Pargo had minor effect on rates of denitrification. Mass balances in both mangroves showed that a large percentage of the added $^{15}\text{NO}_3^-$ was transformed into particulate nitrogen in the sediment, while a small fraction was reduced to NH_4^+ . The dominance of these nitrogen transformations can probably be associated with the low availability of NO_3^- and the high (>20) C:N ratios of these mangrove sediments. Yet, additional work is needed to evaluate the importance of root and algae uptake in competing with denitrifiers for NO_3^- . Results from this study indicate that the capacity for denitrification in oligotrophic mangrove sediments is low. The reduction of NO_3^- in tidal waters in mangroves, particularly at low concentrations, may not necessarily be associated with loss due to denitrification. Thus, studies of nitrogen cycling based on changes in NO_3^- concentration in tidal waters, such as the studies referred to above, may not accurately describe the function of mangroves as a nitrogen sink.

Studies in coastal marine sediments in temperate regions have shown that nitrification is a major source of nitrate for denitrification (Henriksen & Kemp 1988, Seitzinger 1988, 1990), but it is not clear if this is the case for tropical and subtropical coastal ecosystems (Alongi et al. 1992). Nitrifying bacteria have been measured in mangrove sediments, but nitrification rates are generally low at $<0.22 \mu\text{mol g}^{-1} \text{d}^{-1}$ (Iizumi 1986, Shaiful et al. 1986, Alongi et al. 1992). Shaiful (1987) estimated that nitrate reduction (denitrification) has the capacity to consume 89 to 90% of the NO_3^- derived from nitrification. However, direct estimates of coupled nitrification-denitrification in mangroves are needed to evaluate the contribution of this source of NO_3^- to nitrogen loss in mangrove ecosystems. Spatial and temporal patterns of total denitrification in mangrove sediments of Estero Pargo will depend on determinations of coupled nitrification-denitrification rates to complement the low rates of direct denitrification described in this study.

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LITERATURE CITED

- Alongi DM, Boto KG, Robertson AI (1992) Nitrogen and phosphorous cycles. In: Robertson AI, Alongi DM (eds) Tropical mangrove ecosystems, 41, Coastal and estuarine studies. American Geophysical Union, Washington, DC
- Boto KB (1992) Nutrients and mangroves. In: Connell DW, Hawker DH (eds) Pollution in tropical aquatic systems. CRC Press Inc., Boca Raton, p 129-145
- Boto KG, Saffigna P, Clough B (1985) Role of nitrate in nitrogen nutrition of the mangrove *Avicennia marina*. *Mar Ecol Prog Ser* 21:259-265
- Boto KG, Wellington JT (1984) Soil characteristics and nutrient status in a northern Australian mangrove forest. *Estuar coast Shelf Sci* 7:61-69
- Boto KG, Wellington JT (1988) Seasonal variation in concentrations and fluxes of dissolved organic and inorganic materials in a tropical tidally-dominated mangrove waterway. *Mar Ecol Prog Ser* 50:151-160
- Bremner JM (1965) Inorganic forms of nitrogen. In: Black CA et al. (eds) Methods of soil analysis, part 2.9. Am Soc of Agron, Madison, WI, p 1179-1237
- Caffrey JM, Kemp WK (1992) Influence of the submersed plant, *Potamogeton perfoliatus*, on nitrogen cycling in estuarine sediments. *Limnol Oceanogr* 37:1483-1495
- Carlson PR, Yabro LA (1988) Physical and biological control of mangrove pore water chemistry. In: Hook DD et al. (eds) The ecology and management of wetlands, Vol 1, Ecology of wetlands. Timber Press, Portland, OR, p 112-132
- Christensen PB, Nielsen LP, Sørensen J, Revsbech NP (1990) Denitrification in nitrate-rich streams; diurnal and seasonal variation related to benthic oxygen metabolism. *Limnol Oceanogr* 35:640-651
- Cole JA, Brown CM (1980) Nitrite reduction to ammonia by fermentative bacteria: a short circuit in the biological nitrogen cycle. *FEMS Microbiol Lett* 7:65-72
- Corredor JE, Morell MJ (1994) Nitrate depuration of secondary sewage effluents in mangrove sediments. *Estuaries* 17:295-300
- Curran M (1985) Gas movements in the roots of *Avicennia marina* (Forsk.) Vierh. *Austr J Plant Physiol* 12:97-108
- Davidson EA, Hart SC, Firestone MK (1992) Internal cycling of nitrate in soils of a mature coniferous forest. *Ecology* 73:1148-1156
- Day JW, Conner WH, Ley-Lou F, Day RH, Navarro AM (1987) The productivity and composition of mangrove forests, Laguna de Terminos, Mexico. *Aquat Bot* 27:267-284
- Day JW, Day RH, Barreiro MT, Ley-Lou F, Madden CJ (1982) Primary production in the Laguna de Terminos, a tropical estuary in the southern Gulf of Mexico. *Oceanol Acta* 5: 269-276
- Dor I, Levy I (1984) Primary productivity of the benthic algae in the hard-bottom mangal of Sinai. In: Por FD, Dor I (eds) Hydrobiology of the mangal. 20, Development in hydrobiology. Dr W Junk Publishers, The Hague, p 179-191
- Fiedler R, Proksch G (1975) The determination of nitrogen-15

- by emission and mass spectrometry in biochemical analysis: a review. *Anal Chim Acta* 78:1–62
- Grasshoff K, Erhardt M, Kremling K (1983) Methods of seawater analysis. Verlag Chemie, Weinheim
- Hart SC, Nason GE, Myrold DD, Perry DA (1994) Dynamics of gross nitrogen transformation in an old-growth forest: the carbon connection. *Ecology* 75:880–891
- Henriksen K, Kemp WM (1988) Nitrification in estuarine and coastal marine sediments. In: Blackburn TH, Sørensen J (eds) Nitrogen cycling in coastal marine environments, 33. SCOPE, John Wiley and Sons, New York, p 207–249
- Iizumi H (1986) Soil nutrient dynamics. In: Cragg S, Polunin N (eds) Workshop on mangrove ecosystem dynamics. UNDP/UNESCO Regional Project (RAS/79/002), New Delhi, p 171–180
- Jenkins MC, Kemp WM (1984) The coupling of nitrification and denitrification in two estuarine sediments. *Limnol Oceanogr* 29:609–619
- Katyal JC, Holt LS, Gadalla AM (1989) A method to determine soil-entrapped denitrification products in submerged soils. *Soil Sci Soc Am J* 53:296–297
- Keeney DR, Nelson DW (1982) Nitrogen — inorganic forms. In: Page AL, Miller RH, Keeney DR (eds) Methods of soil analysis, Part 2. Chemical and microbiological properties. ASA-SSSA, Madison, WI, p 643–698
- Kessel CV, Pennock DJ, Farrel RE (1993) Seasonal variations in denitrification and nitrous oxide evolution at the landscape scale. *Soil Sci Soc Am J* 57:988–995
- King D, Nedwell DB (1987) The adaptation of nitrate reducing bacterial communities in estuarine sediments in response to overlying nitrate load. *FEMS Microbiol Ecol* 45:15–26
- Koike I, Sørensen J (1988) Nitrate reduction and denitrification in marine sediments. In: Blackburn TH, Sørensen J (eds) Nitrogen cycling in coastal marine environments. John Wiley & Sons, Ltd, New York, p 251–273
- Ley-Lou F (1985) Aquatic primary productivity, nutrient chemistry, and oyster community ecology in a mangrove bordered tidal channel, Laguna de Terminos, Mexico. MSc thesis, Louisiana State University, Baton Rouge
- Lindau CW, DeLaune RD (1991) Dinitrogen and nitrous oxide emission and entrapment in *Spartina alterniflora* salt-marsh soils following addition of N-15 labeled ammonium and nitrate. *Estuar coast Shelf Sci* 32:161–172
- Lindau CW, Patrick WH, DeLaune RD, Reddy KR, Bollich PK (1988) Entrapment of nitrogen-15 dinitrogen during soil denitrification. *Soil Sci Soc Am J* 52:538–540
- Lugo AE (1990) Fringe wetlands. In: Lugo AE, Brinson M, Brown S (eds) Ecosystems of the world: forested wetlands. Elsevier, Amsterdam, p 143–169
- Lugo AE, Brown S, Brinson MM (1990) Concepts in wetland ecology. In: Lugo AE, Brinson M, Brown S (eds) Ecosystems of the world: forested wetlands. Elsevier, Amsterdam, p 53–85
- Lugo AE, Snedaker SC (1974) The ecology of mangroves. *A Rev Ecol Syst* 5:39–64
- Lynch JC (1989) Sedimentation and nutrient accumulation in mangrove ecosystems of the Gulf of Mexico. MSc thesis, The University of Southwestern Louisiana, Lafayette
- Lynch JC, Meriwether JR, McKee BA, Vera-Herrera F, Twilley RR (1989) Recent accretion in mangrove ecosystems based on ¹³⁷Cs and ²¹⁰Pb. *Estuaries* 12:284–299
- Mackin JE, Aller RC (1984) Ammonium adsorption in marine sediments. *Limnol Oceanogr* 29:250–257
- McKee KL (1993) Soil physiochemical patterns and mangrove species distribution — reciprocal effects? *J Ecol* 81:477–487
- McKee KL, Mendelssohn IA, Hester MW (1988) Reexamination of pore water sulfide concentrations and redox potentials near the aerial roots of *Rhizophora mangle* and *Avicennia germinans*. *Am J Bot* 75:1352–1359
- Morris JT (1991) Effects of nitrogen loading on wetland ecosystems with particular reference to atmospheric deposition. *A Rev Ecol Syst* 22:257–279
- Mosier AR, Schimel DS (1993) Nitrification and denitrification. In: Knowles R, Blackburn TH (eds) Nitrogen isotope techniques. Isotopic techniques in plant, soil, and aquatic biology. Academic Press, San Diego, p 181–208
- Mulvaney RL, Boast CW (1986) Equations for determination of nitrogen-15 labeled dinitrogen and nitrous oxide by mass spectrometry. *Soil Sci Soc Am J* 50:360–363
- Mulvaney RL, Kurtz LT (1982) A new method for determination of ¹⁵N-labeled nitrous oxide. *Soil Sci Soc Am J* 46:1178–1184
- Mulvaney RL, Kurtz LT (1984) Evolution of dinitrogen and nitrous oxide from nitrogen-15 fertilized soil cores subjected to wetting and drying cycles. *Soil Sci Soc Am J* 48:596–602
- Nedwell DB (1975) Inorganic nitrogen metabolism in a eutrophicated tropical mangrove estuary. *Water Res* 9:221–231
- Nickerson NH, Thibodeau FR (1985) Association between pore water sulfide concentrations and the distribution of mangroves. *Biogeochem* 1:183–192
- Nielsen LP, Christensen PB, Revsbech NP (1990) Denitrification and photosynthesis in stream sediment studied with microsensor and whole-core techniques. *Limnol Oceanogr* 35:1135–1144
- Nishio T, Koike I, Hattori A (1982) Denitrification, nitrate reduction, and oxygen consumption in coastal and estuarine sediments. *Appl Environ Microbiol* 43:648–653
- Nixon SW, Lee V (1986) Wetlands and water quality: a regional review of recent research in the United States on the role of freshwater and saltwater wetlands as sources, sinks, and transformers of nitrogen, phosphorous, and various heavy metals. Contract No. DACW39–83-M-0366, US Army Engineer Waterways Experiment Station, Vicksburg, MS
- Phleger FB, Ayala-Castañares A (1971) Processes and history of Laguna de Terminos, Mexico. *Bull Am Ass Petrol Geol* 55:2130–2140
- Por FD (1984) The ecosystem of the mangal: general considerations. In: Por FD, Dor I (eds) Hydrobiology of the mangal: the ecosystem of the mangrove forest. 20, Developments in hydrobiology. Dr W Junk Publishers, The Hague, p 1–14
- Reedy KR, Patrick WH, Lindau CW (1989) Nitrification-denitrification at the plant root-sediment interface in wetlands. *Limnol Oceanogr* 34:1004–1013
- Rehr B, Klemme JH (1989) Competition for nitrate between denitrifying *Pseudomonas stutzeri* and nitrate ammonifying enterobacteria. *FEMS Microbiol Ecol* 62:51–64
- Risgaard-Petersen N, Rysgaard S, Nielsen LP, Revsbech NP (1994) Diurnal variation of denitrification and nitrification in sediments colonized by benthic microphytes. *Limnol Oceanogr* 39:573–579
- Rivera-Monroy VH, Day WJ, Twilley RR, Vera-Herrera F, Coronado-Molina C (1995) Flux of nitrogen and sediment in a fringe mangrove forest in Terminos Lagoon, Mexico. *Estuar coast Shelf Sci* 40:139–160
- Rodriguez C, Stoner A (1990) The epiphyte community of mangrove roots in a tropical estuary: distribution and biomass. *Aquat Bot* 36:117–126
- Rosenfeld JK (1979) Ammonium adsorption in nearshore anoxic sediments. *Limnol Oceanogr* 24:356–364
- SAS (1993) JMP® user's guide. SAS Institute, Cary, NC

- Scholander PF, van Dam L, Scholander SI (1955) Gas exchange in the roots of mangroves. *Am J Bot* 42:92–98
- Seitzinger SP (1988) Denitrification in freshwater and coastal marine sediments: ecological and geochemical significance. *Limnol Oceanogr* 33:702–724
- Seitzinger SP (1990) Denitrification in aquatic sediments. In: Revsbech NP, Sørensen J (eds) *Denitrification in soil and sediments*. Plenum, New York, p 301–322
- Shaiful AAA (1987) Nitrate reduction in mangrove swamps. *Malaysian appl Biol* 16:361–367
- Shaiful AAA, Abdul Manan DM, Ramli MR, Veerasamy R (1986) Ammonification and nitrification in wet mangrove soils. *Malays J Sci* 8:47–56
- Smith CJ, Delaune RD, Patrick WH (1982) Nitrate reduction in *Spartina alterniflora* marsh soil. *Soil Sci Soc Am J* 46:748–750
- Solórzano L (1969) Determination of ammonia in natural waters by the phenylhypochlorite method. *Limnol Oceanogr* 14:799–801
- Sørensen J (1978) Capacity for denitrification and reduction of nitrate to ammonia in a coastal marine sediment. *Appl environ Microbiol* 35:301–305
- Steyer GD (1988) Litter dynamics and nitrogen retranslocation in three types of mangrove forests in Rookery Bay, Florida. MSc thesis, The University of Southwestern Louisiana, Lafayette
- Sumi T, Koike I (1990) Estimation of ammonification and ammonium assimilation in surficial coastal and estuarine sediments. *Limnol Oceanogr* 35:270–286
- Thibodeau FR, Nickerson NH (1986) Differential oxidation of mangrove substrate by *Avicennia germinans* and *Rhizophora mangle*. *Am J Bot* 73:512–516
- Twilley RR (1988) Coupling of mangroves to the productivity of estuarine and coastal waters. In: Jansson BO (ed) *Coastal-offshore ecosystems: interactions*. 22, Lecture notes on coastal and estuarine studies. Springer-Verlag, Berlin, p 80–155
- Twilley RR, Chen RH, Hargis T (1992) Carbon sinks in mangroves and their implications to carbon budget of tropical ecosystems. *Water Air Soil Pollut* 64:265–288
- Twilley RR, Kemp WM (1986) Factors controlling the variability of denitrification potentials in Chesapeake Bay sediments. In: Wolfe DA (ed) *Estuarine variability*. Academic Press, Orlando, p 277–293
- Twilley RR, Lugo AE, Patterson-Zucca C (1986) Production, standing crop, and decomposition of litter in basin mangrove forests in southwest Florida. *Ecology* 67:67–683
- Twilley RR, Snedaker SC, Yañez-Arancibia A, Medina E (1995) Biodiversity and ecosystem processes in tropical estuaries: perspectives from mangrove ecosystems. In: Mooney H, Cushman H, Medina E (eds) *Biodiversity and ecosystem functions: a global perspective*. John Wiley and Sons, New York (in press)
- Walsh GE (1967) An ecological study of a Hawaiian mangrove swamp. In: Lauff GH (ed) *Estuaries*. American Association for the Advancement of Science, Washington, DC, p 420–431
- Weiss RF (1970) The solubility of nitrogen, oxygen and argon in water and seawater. *Deep Sea Res* 17:721–735
- Yañez-Arancibia A, Day JW (1988) Ecological characterization of the Terminos Lagoon. In: Yañez-Arancibia A, Day JW (eds) *Ecology of coastal ecosystems in the Southern Gulf of México: the Terminos Lagoon region*. UNAM Press, Mexico, p 1–26

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