MANGROVE ISOTOPIC (δ^{15}N AND δ^{13}C) FRACTIONATION ACROSS A NITROGEN VS. PHOSPHORUS LIMITATION GRADIENT

Karen L. McKee,[1] Ilka C. Feller,[2] Marianne Popp,[3] and Wolfgang Wanek[3]

[1]*National Wetlands Research Center, U.S. Geological Survey, 700 Cajundome Boulevard, Lafayette, Louisiana 70506 USA*
[2]*Smithsonian Environmental Research Center, P.O. Box 28, Edgewater, Maryland 21037 USA*
[3]*Institute of Plant Physiology, University of Vienna, Vienna, Austria*

Abstract. Mangrove islands in Belize are characterized by a unique switching from nitrogen (N) to phosphorus (P) limitation to tree growth from shoreline to interior. Fertilization has previously shown that *Rhizophora mangle* (red mangrove) fringe trees (5–6 m tall) growing along the shoreline are N limited; dwarf trees ($\leq$1.5 m tall) in the forest interior are P limited; and transition trees (2–4 m tall) are co-limited by both N and P. Growth patterns paralleled a landward decrease in soil flushing by tides and an increase in bioavailable N, but P availability remained consistently low across the gradient. Stable isotopic composition was measured in *R. mangle* leaves to aid in explaining this nutrient switching pattern and growth variation. Along control transects, leaf δ^{15}N decreased from +0.10‰ (fringe) to −5.38‰ (dwarf). The δ^{15}N of N-fertilized trees also varied spatially, but the values were consistently more negative (by ∼3‰) compared to control trees. Spatial variation in δ^{15}N values disappeared when the trees were fertilized with P, and values averaged +0.12‰, similar to that in control fringe trees. Neither variation in source inputs nor microbial fractionation could fully account for the observed patterns in δ^{15}N. The results instead suggest that the lower δ^{15}N values in transition and dwarf control trees were due to plant fractionation as a consequence of slower growth and lower N demand. P fertilization increased N demand and decreased fractionation. Although leaf δ^{13}C was unaffected by fertilization, values increased from fringe (−28.6‰) to transition (−27.9‰) to dwarf (−26.4‰) zones, indicating spatial variation in environmental stresses affecting stomatal conductance or carboxylation. The results thus suggest an interaction of external supply, internal demand, and plant ability to acquire nutrients under different hydro-edaphic conditions that vary across this tree-height gradient. The findings not only aid in understanding mangrove discrimination of nitrogen and carbon isotopes, but also have implications for identifying nutrient loading and other stress conditions in coastal systems dominated by mangroves.

Key words: carbon isotope discrimination; height forms; mangrove, red; nitrogen isotope discrimination; nutrient limitation; Rhizophora mangle; stress tolerance.

INTRODUCTION

Variation in plant height and productivity across environmental gradients occurs in both marsh and mangrove ecosystems. Along the Atlantic coast, tall creekbank and short inland height forms of *Spartina alterniflora* are characteristic features of intertidal marshes (Mendelssohn 1979). At subtropical and tropical latitudes, mangrove wetlands also exhibit tree-height and productivity variation from tall, highly productive trees growing along shorelines or riverbanks to scrub or dwarf stands in interior forests (Lugo and Snedaker 1974, Koltes et al. 1998). An understanding of the determinants of primary productivity in marsh and mangrove ecosystems is important because of its relationship to productivity of coastal fisheries (Turner 1977, Primavera 1998). Work conducted in temperate salt marshes indicates that local variation in *S. alterniflora* growth is mainly controlled by abiotic factors, specif-

ically the interaction of soil anoxia, soluble sulfide, and salinity with plant nitrogen (N) uptake and assimilation (Mendelssohn 1979, Morris 1984, Bradley and Morris 1990, Koch et al. 1990). Investigations into mechanisms controlling mangrove growth suggest that similar edaphic factors interact with plant physiological and morphological adaptations to limit growth across environmental gradients (Boto and Wellington 1984, Ball 1988, Feller 1995, McKee 1995).

In Belize, a complex gradient of stress factors (salinity, flooding, and nutrients) occurs across mangrove-dominated islands where *Rhizophora mangle* L. trees decrease in height from 5–6 m along the shoreline (fringe) to 1.5 m or less in interior dwarf stands (McKee 1995). Productivity, measured as leaf-fall rates, also decreases spatially: fringe = 700 g·m^{-2}·yr^{-1}, transition = 450 g·m^{-2}·yr^{-1}, scrub/dwarf = 280 g·m^{-2}·yr^{-1} (Koltes et al. 1998). The short stature and low productivity of scrub/dwarf trees cannot be attributed to immaturity, since stunted trees are >50 yr old (based on leaf scar counts and leaf production rates; Feller 1995). Feller

TABLE 1. Summary of *Rhizophora mangle* growth responses (percentage increase in growth variable) in three zones (fringe, transition, and dwarf) to nutrient enrichment with either nitrogen (N) as urea or phosphorus (P) as P_2O_5 (from Feller et al. 2001).

Nutrient	Shoot elongation	New leaf pairs	New shoots
Fringe			
N	115	43	233
P	NS	NS	NS
Transition			
N	889	165	900
P	1747	290	1200
Dwarf			
N	NS	NS	NS
P	2579	379	1100

Notes: Increase in growth (relative to controls that received no fertilizer) is based on measurements conducted over a 6-mo interval on replicate trees ($n = 9$ trees per treatment $\times$ zone combination); NS = no significant change from control.

(1995) has also demonstrated that fertilization of dwarf trees with phosphorus (P) can completely reverse this growth limitation. The spatial decline in mangrove tree height does not appear to be due to decreased availability of P to support growth, however. Porewater concentrations of PO_4-P do not vary with distance from the shoreline, and NH_4-N increases (McKee 1993). The co-occurrence of other stressors (e.g., soil waterlogging or salinity) instead suggests that the nutrient limitation to mangroves in landward positions is secondary to other growth-limiting factors, as found for salt marshes (Mendelssohn 1979).

Fertilization of *R. mangle* trees across the entire gradient in Belize has further revealed a unique switching from N to P limitation of mangrove growth. Growth and tissue nutrient responses to fertilization are reported in detail elsewhere (Feller et al. 2001) and summarized in Table 1. Fringe trees responded to N (urea) addition, dwarf trees responded to P addition, and transition trees responded to both N and P (applied separately). Feller et al. hypothesized that this switch from N to P limitation could be due to spatial variation in (1) external supply of N relative to P, (2) soil nutrient transformations affecting relative availability of N and P, and/or (3) nonresource factors affecting plant nutrient uptake, assimilation, and/or translocation.

Stable isotope methodology provides a means to assess nutrient sources (Evans et al. 1996, Rolston et al. 1996, Handley and Scrimgeour 1997), biological transformations affecting nutrient availability (Nadelhoffer and Fry 1994, Nadelhoffer et al. 1996, Jordan et al. 1997), and effects of environmental conditions on plant metabolism and growth (Farquhar 1993, Montoya and McCarthy 1995, Yoneyama 1995). Some evidence from terrestrial plant communities suggests that increased N mineralization rates may lead to increased ^{15}N contents in plant-available N pools and hence higher $\delta^{15}N$ values

in plants. For example, P fertilization of tussock tundra resulted in increased soil N availability and increased $\delta^{15}N$ values in some plant species (Nadelhoffer et al. 1996). Work with mangroves has also demonstrated variation in N and C isotopic composition in association with environmental change (e.g., eutrophication [Fry et al. 2000] and increased salinity [Lin and Sternberg 1992*a, b*]). Fry et al. (2000) reported that $\delta^{15}N$ signatures of *R. mangle* trees varied substantially across sites in south Florida (from $-5\permil$ to $+13\permil$), and concluded that this pattern could be due to variation in source $\delta^{15}N$ (e.g., anthropogenic NO_3^- inputs) and/or to physiological fractionation by plants growing under different stress conditions. Lin and Sternberg (1992*a, b*), studying fringe and scrub *R. mangle* in south Florida, found a negative correlation between leaf carbon isotopes ($\delta^{13}C$) and tree height, and attributed the higher $\delta^{13}C$ values in scrub trees to elevated salinity and consequent effects on stomatal conductance.

In this study, we compared $\delta^{15}N$ and $\delta^{13}C$ values of N- and P-fertilized trees with that of unfertilized trees in an effort to understand the mechanism(s) controlling growth limitations and nutrient switching by *R. mangle* across a tree-height gradient. The results provide insight into within-stand nutrient dynamics in these mangrove ecosystems and factors contributing to isotopic composition of mangrove tissues.

MATERIALS AND METHODS

Site description

The study was conducted at Twin Cays, a peat-based, 92-ha archipelago of mangrove islands located in the Belizean Barrier Reef Complex. These intertidal islands are $\sim$12 km from the mainland and receive no terrigenous inputs of freshwater or sediments. Twin Cays is dominated by *Rhizophora mangle* L. (red mangrove) and *Avicennia germinans* (L.) Stearn. (black mangrove) with scattered *Laguncularia racemosa* (L.) Gaertn. f. (white mangrove). A distinct vegetative pattern is a tree-height gradient that parallels other gradients, such as productivity and tidal fluctuation. A narrow fringe zone (5–20 m wide) of uniformly tall trees occurs in the low intertidal zone around the periphery of the islands. Tree height decreases landward through a transition zone (5–30 m wide) and a dwarf zone (10–30 m wide) in the island interior. Soil surface elevation also varies along this height gradient, with the transition zone being the highest and the dwarf zone being the lowest. Both fringe and dwarf zones are dominated by *R. mangle*, whereas the transition zone is a mixed stand of *R. mangle*, *A. germinans*, and *L. racemosa*. The fringe zone (canopy height = 4–7 m) is flooded and drained >700 times/yr. The transition zone (canopy height = 2–4 m) is flooded only during high spring tides and storms (<50 times/yr). The dwarf zone

(canopy height <2.0 m) is perennially flooded, except during unusually low tides.

Our use of the term ''dwarf'' does not imply a genetic subtype of *R. mangle*, but refers to trees ≤1.5 m in height and is consistent with terminology used in previous work at this location (Feller 1995, Feller et al. 1999, 2001). These stunted trees are mature (i.e., reproductive), and leaf scar counts and leaf production rates indicate that they are >40 yr old (Feller 1995). The lack of a genetic or developmental component has been demonstrated by growing short and tall mangroves under common conditions (Lin and Sternberg 1992b).

Experimental design

Three transects, 10 meters apart, were established at three different locations (*n* = 9) at Twin Cays in January 1995. The transects were oriented perpendicular to the shoreline and traversed the tree-height gradient from shoreline to island interior. Transects were subdivided into three zones based on tree height, and three experimental trees were selected within each zone for a total of nine trees across each transect and a total of 81 trees overall. Each of three transects at each location was randomly assigned to a nutrient treatment (N, P, or control). *Rhizophora mangle* trees were each fertilized at 6-mo intervals with 300 g of N fertilizer as urea (45:0:0), or P fertilizer as P_2O_5 (0:45:0), as described in Feller (1995). Fertilizer was inserted into holes cored in the soil beneath the canopy edge of each tree, and the holes were plugged with peat. At control trees, holes were cored and sealed, but no fertilizer was added.

Hydro-edaphic analyses

To characterize hydro-edaphic conditions across the tree-height gradient, water levels, soil redox potentials and porewater salinity, pH, and sulfide were measured adjacent to the experimental trees in January 1995, before initiation of fertilizer treatments, and again in January 1997 after two years of fertilizer treatment. Water level relative to the soil surface was measured at high and low tides in wells (7 cm in diameter × 30 cm deep) to determine relative depths of inundation and soil flushing over a tidal cycle. Soil redox potentials at 1 cm and 15 cm depths were measured with bright platinum electrodes equilibrated for 30 min as described in McKee et al. (1988). Interstitial water was collected from a depth of 15 cm, and sulfide, pH, and salinity were determined as described previously (McKee et al. 1988).

In January 1998, after three years of fertilizer treatment, bioavailable N and P were assessed with ion-exchange resin bags (Lajtha 1988) placed near each experimental tree. In contrast with instantaneous measures of porewater or extractable N or P, determination of bioavailable N and P with resin bags provides an integrated index of NO_3^-, NH_4^+, and PO_4^{-3} concentra-

tions, the delivery rate (i.e., by diffusion or mass flow), and plant uptake over a time interval. The bags were constructed of undyed, nylon mesh material (panty hose), filled with 4 g of a mixed-bed (H^+/OH^-) exchange resin (16–50 mesh) and conditioned with 1 mol/L NaCl. After rinsing with deionized water, duplicate bags were inserted into 15-cm-deep holes at each sampling station and covered with a soil plug. After incubation in situ for 15 d, the bags were retrieved, desorbed with 0.5 mol/L HCl (PO_4^{-3} and NO_3^-) or 1 mol/L KCl (NH_4^+), and the extract analyzed with a LACH-AT system (Quikchem IV, Lachat Instruments, Milwaukee, Wisconsin, USA) for NH_4^+, NO_3^-, and PO_4^{-3}. Unincubated resin bags were used as blanks.

Plant and peat analyses

Leaf samples for isotopic analysis were harvested from each of 81 experimental trees in January 1998. The youngest, fully mature green leaves were collected from a penapical stem position on first-order branches in the top of the canopy (sun leaves). Peat cores (5 cm in diameter × 20 cm deep) were also collected from unenriched zones to determine isotopic composition of soil organic matter. Leaf and peat samples were frozen and then freeze dried within 48 h of collection. The stable isotope composition ($\delta^{13}C$ and $\delta^{15}N$ signatures) was determined at the Institute of Plant Physiology, University of Vienna, Austria. Samples were ground to a fine powder in a ball mill (Retsch MM2, Vienna, Austria) and analyzed by continuous-flow gas isotope ratio mass spectrometry. The elemental analyzer (EA 1110, CE Instruments, Milan, Italy) was interfaced via a ConFlo II device (Finnigan MAT, Bremen, Germany) to the gas isotope ratio mass spectrometer (Delta^{Plus}, Finnigan MAT, Bremen, Germany). The ^{13}C and ^{15}N abundances were calculated as follows:

$$\delta^{13}C = ([R_{sample}/R_{standard}] - 1)(1000 \text{ [‰ vs. V-PDB]})$$

$$\delta^{15}N = ([R_{sample}/R_{standard}] - 1)(1000 \text{ [‰ vs. at-air]})$$

where R is the ratio of $^{13}C:^{12}C$ and $^{15}N:^{14}N$, respectively. The standard deviation of repeated measurements of $\delta^{13}C$ and $\delta^{15}N$ values of a laboratory standard was calculated to be 0.10‰ vs. V-PDB and 0.15‰ vs. at-air, respectively. These abundances are referred to the following certified reference materials: Pee Dee Belemnite (PDB) and atmospheric dinitrogen (at-air).

Leaves were also collected from each tree for determination of N and P concentrations and leaf area. Leaf area was determined with a Li-Cor 3000 Portable Area Meter (Li-Cor, Lincoln, Nebraska, USA). Leaf samples were dried at 70°C in a convection oven and ground in a Wiley Mill to pass through a 0.38-mm (40-mesh) screen. Concentration of N was determined with a 2400 CHN Analyzer (Perkin-Elmer, Shelton, Connecticut, USA) at the Smithsonian Environmental Research Center, Edgewater, Maryland, USA. Concentration of P was determined after acid digestion by using

TABLE 2. Summary of hydro-edaphic conditions measured at Twin Cays prior to fertilizer application across a *Rhizophora mangle* tree-height gradient.

Variable	Fringe	Transition	Dwarf	P
Water depth relative to soil surface (cm)				
High tide	+3.2 ± 0.6	+2.8 ± 0.6	+12.4 ± 3.1	0.0015
Low tide	−15.0 ± 1.9	−8.1 ± 2.0	+0.9 ± 2.4	<0.0001
Soil E_h (mV)				
1 cm depth	50 ± 15	61 ± 16	−9 ± 13	
	(−42 to +20)	(42 to 102)	(−97 to −6)	<0.0001
15 cm depth	−17 ± 11	−16 ± 15	−80 ± 11	
	(−76 to −9)	(−46 to −34)	(−177 to −94)	<0.0001
Porewater				
Salinity (g/L)	37 ± 1	39 ± 1	40 ± 1	
	(34–39)	(37–67)	(35–39)	
pH	6.3 ± 0.1	6.3 ± 0.1	6.4 ± 0.1	0.0002
	(6.5–6.6)	(6.0–6.7)	(6.6–6.9)	NS
Sulfide (mmol/L)	0.36 ± 0.05	0.43 ± 0.06	0.73 ± 0.10	
	(0.08–0.20)	(0.01–0.27)	(0.64–1.68)	0.0001

Notes: Values are the mean (± 1 SE; $n = 9$ for water depth and $n = 27$ for soil and porewater variables). Probability of F test results of an ANOVA are given for main effect of zone (df = 2, 68). Seasonal ranges (in parentheses) of physico-chemical variables at Twin Cays are also given for comparison (from McKee 1993*a*).

an inductively coupled plasma (ICP) spectrometer at Analytical Services, Pennsylvania State University, State College, Pennsylvania, USA.

Statistical analyses

The plant isotope data were analyzed by an analysis of variance (ANOVA, fixed effects model) in which the grouping factors were fertilizer treatment (control, N, and P) and zone (fringe, transition, and dwarf). Environmental data were analyzed with a repeated-measures ANOVA (fixed-effects model) where the grouping factors were treatment, zone, and sampling date (January 1995 and 1997). Any data that did not meet the variance homogeneity or normality assumptions for ANOVA were transformed and re-tested prior to analysis. However untransformed means (±1 SE) are presented. Relationships among plant and edaphic variables were examined by correlation analysis.

RESULTS

Along experimental transects, the dwarf zone was inundated to a greater depth at high tide, and the water level remained above the soil surface over a diurnal tidal cycle compared to the fringe and transition zones where greater soil flushing occurred (Table 2). Soil physico-chemical factors associated with flooding (E_h and sulfide concentrations) also varied and reflected the greater inundation and infrequent soil flushing of the dwarf zone (Table 2). Salinity was significantly elevated in the transition and dwarf zones in January prior to fertilization (Table 2). Fertilization with N or P did not significantly alter salinity, pH, or E_h (data not shown), but N enrichment significantly increased sulfide concentrations in the fringe zone to 0.68 ± 0.11

mmol/L, compared to 0.08 ± 0.04 mmol/L and 0.07 ± 0.03 mmol/L along control and P-fertilized transects, respectively (date × treatment × zone interaction; $F_{4, 126} = 2.399$, $P = 0.05$).

Bioavailable NH_4-N increased from the fringe to the dwarf zone, but NO_3-N and PO_4-P did not vary (Table 3). Concentrations of NO_3-N were frequently below detection limits, so values for NO_3-N and NH_4-N were added to compare N bioavailability (Fig. 1). Across control transects, the ratio of bioavailable N:P in fringe and transition zones was significantly lower than in the dwarf zone (1 df contrast: $t = 2.83$, $P \leq 0.01$; Fig. 1). Fertilization with N or P caused significant increases in bioavailability of NH_4-N or PO_4-P, respectively (Table 3). Compared to control transects with an N:P ratio of 17 (averaged over zone), fertilization with N increased the ratio to 23, whereas P addition decreased it to 5.0 ($F_{2,68} = 6.47$, $P \leq 0.001$; Fig. 1).

Stable isotope values ($\delta^{15}N$) of source inputs are summarized in Table 4. The $\delta^{15}N$ values for mangrove peat were lower in the dwarf zone (1 df contrast; $P = 0.0159$) compared to the fringe and transition zones, which were not significantly different (Table 5). The $\delta^{13}C$ values did not differ across zones and averaged −26.5 ± 0.16 (Table 5). The C and N concentrations of peat averaged 310 mg/g and 13.7 mg/g, respectively, and showed slight differences across zones (Table 5). The C:N ratios of mangrove peat decreased from fringe and transition to dwarf zones, but the difference was not significant (Table 5).

Leaf $\delta^{15}N$ values decreased across the control transect from +0.1‰ in the fringe to −5.38‰ in the dwarf zone (Fig. 2a). The $\delta^{15}N$ signatures of N-fertilized trees also varied spatially, but the values were consistently

TABLE 3. Summary of ANOVA results for bioavailable NO_3-N, NH_4-N, and PO_4-P.

Bioavailable nutrient	Zone					Treatment				
	Mean (SE)					Mean (SE)				
	Fringe	Transition	Dwarf	F	P	Control	N	P	F	P
NO_3-N	0.016 (0.008)	0.006 (0.003)	0.003 (0.002)	2.18	NS	0.002 (0.002)	0.011 (0.005)	0.012 (0.007)	1.23	NS
NH_4-N	0.088 (0.025)	0.110 (0.028)	0.205 (0.029)	5.29	**	0.114 (0.025)	0.210 (0.038)	0.088 (0.014)	5.71	**
NO_3^-:NH_4^+	0.591 (0.320)	0.097 (0.053)	0.031 (0.024)	3.15	*	0.020 (0.019)	0.145 (0.092)	0.515 (0.290)	2.57	NS
NO_3^- + NH_4^+	0.102 (0.025)	0.115 (0.027)	0.209 (0.029)	4.60	**	0.117 (0.025)	0.218 (0.037)	0.099 (0.014)	5.71	**
PO_4-P	0.017 (0.003)	0.019 (0.006)	0.018 (0.003)	0.057	NS	0.011 (0.002)	0.013 (0.002)	0.030 (0.006)	6.39	**
N : P	24 (6)	26 (7)	51 (11)	3.91	*	38 (8)	51 (11)	12 (2)	6.47	**

Note: Nutrients were measured with resin bags implanted in fringe, transition, and dwarf zones and fertilizer treatments ($\mu g \cdot [g\ resin]^{-1} \cdot d^{-1}$; mean $\pm$ 1 SE, $n = 9$).
  * $P \leq 0.05$, **$P \leq 0.01$.

more negative (from $-3‰$ to $-8‰$) compared to the control trees in each respective zone (Fig. 2a). This spatial variation in $\delta^{15}N$ values disappeared when the trees were fertilized with P, however, and averaged $+0.12‰$ across zones, similar to that of fringe control trees (Fig. 2a). There was a significant increase in $\delta^{13}C$ values from the fringe ($-28.4‰$) to transition ($-27.5‰$) and dwarf ($-26.5‰$) zones that varied little by fertilizer treatment (Fig. 2b).

Along the control transects, $\delta^{15}N$ values showed a

FIG. 1. Relative bioavailability of nitrogen (NO_3^- + NH_4^+) and phosphorus (PO_4^{-3}) measured with resin bags across the tree-height gradient. Symbols represent three tree-height zones: fringe, 5–6 m tall (squares); transition, 3–4 m tall (diamonds); and dwarf, $\leq$1.5 m tall (circles). Three treatments are also represented: controls, no fertilizer added (open symbols); phosphorus added as P_2O_5 (solid symbols); nitrogen added as urea (cross symbols). Values are the mean $\pm$ 1 SE ($n = 9$); note that all SE bars are plotted.

strong negative correlation with $\delta^{13}C$ values ($r = 0.84$, $F_{1,25} = 59.7870$, $P < 0.0001$, $y = -28.3027 - 0.3225x$). There was also a negative correlation between $\delta^{15}N$ and $\delta^{13}C$ signatures along the N-fertilized transect ($r = 0.71$, $F_{1,25} = 24.5018$, $P < 0.0001$, $y = -28.9467 - 0.2956x$). However P fertilizer dramatically altered the relationship between $\delta^{15}N$ and $\delta^{13}C$ ($r = 0.28$, $F_{1,25} = 2.2613$, $P = 0.1452$, $y = -27.7967 - 0.2850x$). Leaf $\delta^{15}N$ values showed a strong negative correlation with leaf N:P ($r = 0.968$, $F = 103.99$, $P < 0.0001$; $y = -13.55 + 195.9(1/x)$; Fig. 3). There was a significant positive correlation between leaf $\delta^{13}C$ values and leaf N on a unit area basis ($r = 0.877$, $F = 23.47$, $P = 0.0019$; $y = -23.049 - 1.00(1/x)$; Fig. 4). Leaf P (but not leaf N) was negatively correlated with water depth at low tide ($r = 0.707$, $F = 7.00$, $P = 0.0331$; $1/y = 2.35 + 0.039x$; Fig. 5).

DISCUSSION

Hydro-edaphic conditions

Consistent with previous findings (McKee 1993, 1995), hydro-edaphic conditions varied across the tree-height gradient and indicated less soil flushing and lower redox status with accumulation of sulfide in the dwarf zone (Table 2). The higher salinity in transition and dwarf mangrove zones also reflects infrequent tidal flushing and accumulation of salts (Table 2). Other work conducted at these sites found higher salinity during the dry season (McKee 1993, 1995). Salinity in the transition zone may vary from sea strength to >80 g/L, whereas it fluctuates less in the fringe and dwarf zones and usually does not exceed 40–45 g/L.

Bioavailability of N increased relative to P across the gradient (Fig. 1, Table 3) and agreed with growth responses to fertilization (Table 1). Similarly, pore-water NH_4^+ in salt marshes may be ten times higher in less productive inland zones compared to streamside (Mendelssohn 1979). The higher NH_4^+ in interior
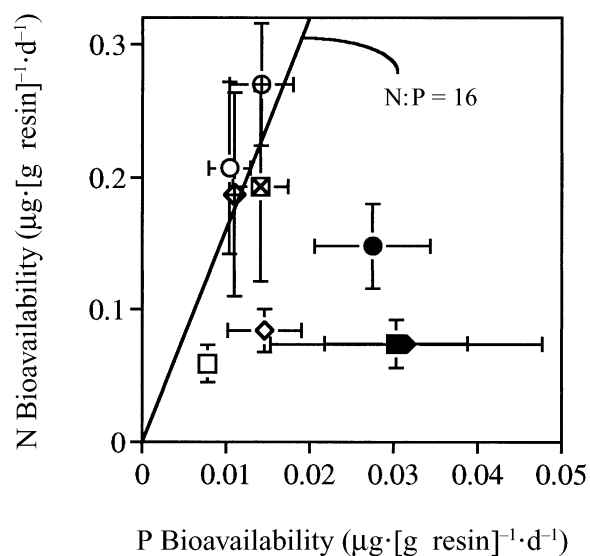
TABLE 4. Values of $\delta^{15}N$ (‰) for nitrogen (N) source inputs.

N Source	$\delta^{15}N$	Reference
Oceanic NO_3^-	+4 to +7	Macko and Ostrom (1994)
Fertilizer NO_3^-	−3 to +2	Rolston et al. (1996)
Animal waste	+9 to +25	Rolston et al. (1996)
Soil organic N	+2 to +8	Rolston et al. (1996)
Mangrove peat	−0.13	this study
Root epibionts	+5 to +6	Ellison et al. (1996)
N fixation	−1 to +1.5	Carpenter et al. (1997)
Urea fertilizer	−0.34	this study

marsh or mangrove zones could be the result of higher inputs (e.g., nitrogen fixation and litter mineralization), lower losses (e.g., denitrification), and/or decreased plant uptake (e.g., due to slower plant growth). Boto and Wellington (1984) also found a similar spatial variation in N:P that suggested N limitation to Australian mangroves at lower intertidal positions and P limitation at higher intertidal (inland) positions. In contrast to Twin Cays, however, P availability varied spatially in Australian mangal (Boto and Wellington 1983). This was possibly due to decreased inputs of mineral sediment with distance from the shoreline or to co-precipitation with Fe, a redox couple, (e.g., in more oxidized soils at higher elevations). Since the majority of P at Twin Cays likely occurs in organic form (peat soil with >75% organic matter) or as Ca-bound compounds (calcium carbonate sand), soil E_h would not be expected to strongly influence P availability, as found in Australia.

Thus the tree-height gradient at Twin Cays was characterized by variation in soil waterlogging, salinity, and nutrient availability, analogous to that described for salt marshes where uptake and assimilation of N is limited by flooding-related stresses (Mendelssohn 1979, Morris 1984, Bradley and Morris 1990). However hydroedaphic patterns (Tables 2 and 3, Fig. 1), in combination with the nutrient-switching limitation to plant growth (Table 1), suggest a more complex interaction between availability of N and P and growth-limiting stress factors in these mangrove forests.

Nitrogen isotope composition

The range of *R. mangle* leaf $\delta^{15}N$ measured across the tree-height gradient in Belize (Fig. 2) and in south

Florida (Fry et al. 2000) was wider than that reported for some terrestrial plant species across greater spatial distances (≤ 2.5‰; Nadelhoffer et al. 1996). The broad range of values found for mangrove height forms in close proximity suggests strong spatial variation in processes controlling discrimination of N isotopes. Several factors may affect $\delta^{15}N$ signatures, including source of N, microbial processes that deplete or enrich ^{15}N in soil or water, and plant physiological processes that discriminate against ^{15}N. The dramatic change in $\delta^{15}N$ values across the control transects suggests that one or more of these factors is important in determining the switch from N to P limitation.

The simplest explanation for spatial change in isotopic values is variation in source ^{15}N. With increasing distance from the shoreline, the plants could be utilizing other sources with more negative signatures. Isotopic analyses by Ellison et al. (1996) suggest that fringing *R. mangle* trees at Twin Cays may acquire inorganic N from sponges encrusting the prop roots submerged in tidal creeks. The epibiont sources had signatures that varied from +5‰ to +6‰ (Table 4), and associated mangrove leaves exhibited $\delta^{15}N$ values between zero and +0.5‰ (Ellison et al. 1996), similar to our control fringe trees (Fig. 2). Another source of N is that fixed by nitrogen-fixing organisms such as cyanobacteria (Potts 1979). Fertilization with P may stimulate N fixation and increase foliar $\delta^{15}N$, as found for a tree species (*Metrosideros polymorpha*) growing on volcanic substrate (Vitousek 1999). Although variation in source N may have contributed to the observed isotopic patterns, this scenario by itself does not explain the spatial variation in tree height nor the switch

TABLE 5. Mean isotopic (‰) and chemical composition, and ANOVA results, for mangrove peat cores (20 cm depth), collected in unenriched fringe, transition, and dwarf forest zones at Twin Cays.

	Zone			ANOVA	
Variable	Fringe	Transition	Dwarf	F	P
$\delta^{15}N$	+0.13 ± 0.25	+0.03 ± 0.07	−0.53 ± 0.05	5.625	0.0421
$\delta^{13}C$	−26.42 ± 0.14	−26.54 ± 0.08	−26.53 ± 0.08	0.3973	NS
N (mg/g)	13.7 ± 0.4	13.1 ± 0.1	14.2 ± 0.3	3.754	0.0876
C (mg/g)	322 ± 9	314 ± 9	293 ± 2	3.584	0.0948
C : N	27.5 ± 0.3	28.0 ± 0.7	24.0 ± 0.3	4.025	0.0779

Note: Mean values are reported ± 1 SE; $n = 3$.
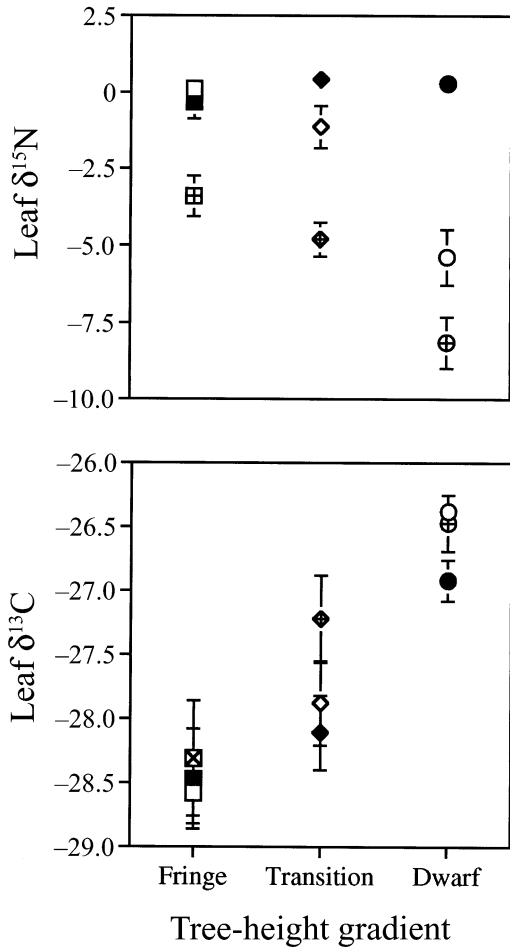
FIG. 2. Variation in $\delta^{15}N$ and $\delta^{13}C$ values (‰) measured in *Rhizophora mangle* leaves across a tree-height gradient in response to spatial position (zone) and fertilization. Symbols are as in Fig. 1. Values are means ± 1 SE ($n = 9$); note that all SE bars are plotted.

FIG. 3. Variation in $\delta^{15}N$ values (‰) and leaf nitrogen (N) : phosphorus (P) ratios measured in *Rhizophora mangle* across a tree-height gradient in response to spatial position (zone) and fertilization. Symbols as in Fig. 1. Values are means ± 1 SE ($n = 9$); note that all SE bars are plotted.

losses of N or uncoupled reactions. This model is consistent with the landward decline in soil flushing/aeration and leaf $\delta^{15}N$ values across the control transect as well as the higher availability of NH_4^+ near dwarf trees (Tables 2 and 3), but does not explain the effect of P fertilization (Fig. 2).

Plants may also discriminate against $\delta^{15}N$ during uptake, assimilation, and/or translocation. Where N is limiting, all N is typically used regardless of isotope (Montoya and McCarthy 1995, Evans et al. 1996). Since the fringe trees are N limited (Table 1), they

from N to P limitation to mangrove growth across the gradient.

Leaf $\delta^{15}N$ may also be altered indirectly by fractionation processes (microbial or plant). Fry et al. (2000) proposed two models to explain interactions of isotope source and processing to generate isotopic gradients across mangrove stands: (1) a source of low $\delta^{15}N$ value with higher plant $\delta^{15}N$ due to microbial fractionation of N isotopes, and (2) a source of high $\delta^{15}N$ value with lower plant $\delta^{15}N$ due to plant isotopic fractionation during N uptake. We consider these two models further in explaining the results at Twin Cays.

Microbial fractionation may cause enrichment with ^{15}N due to losses of ^{15}N-depleted nitrogen during denitrification or ammonia volatilization (Jordan et al. 1997). Coupled nitrification–denitrification may be greater in the fringe zone, where the soil is periodically flooded and drained by the tides. The dwarf zone, which is almost constantly submerged, may experience lower
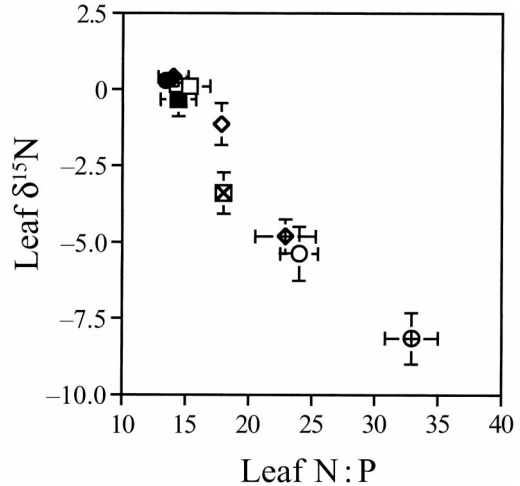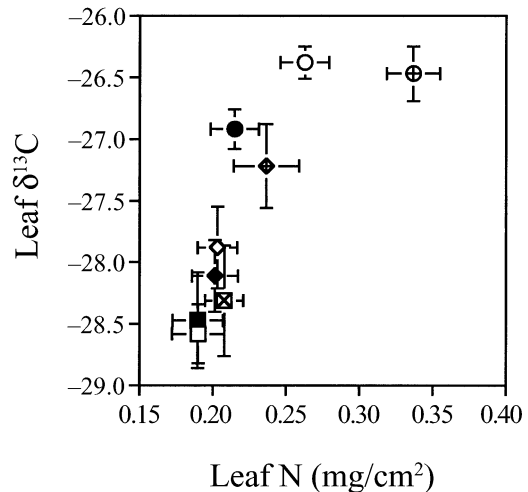

FIG. 4. Variation in leaf $\delta^{13}C$ values (‰) and leaf nitrogen (N) concentration (mg/cm^2) measured in *Rhizophora mangle* leaves across a tree-height gradient in response to spatial position (zone) and fertilization. Symbols are as in Fig. 1. Values are means ± 1 SE ($n = 9$); note that all SE bars are plotted.
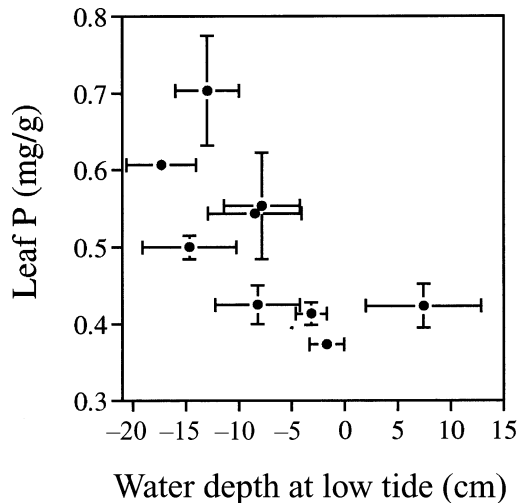
FIG. 5. Variation in leaf phosphorus (P) concentration (mg/g) in control trees, and water depth at low tide (relative to the soil surface) measured at nine stations across the tree-height gradient. Values are means ± 1 SE ($n = 3$); note that all SE bars are plotted.

should use all sources of N, resulting in higher $\delta^{15}N$ values (Fig. 2). Increasing availability of N would result in greater fractionation and lower plant $\delta^{15}N$, as seen with increasing distance from the shoreline as well as in the N-fertilized trees (Fig. 2). Greater plant fractionation also occurs with NH_4^+ than with NO_3^- (Yoneyama et al. 1991, Pennock et al. 1996), so that the decline in foliar $\delta^{15}N$ values with increasing distance from the shoreline might be due to variation in relative availability of these ions. Along the control transect, however, the bioavailability ratio of NO_3^-: NH_4^+ did not vary between fringe and interior zones (1 df contrast: $t = -0.065$, $P > 0.10$). Enrichment with P did result in an increase in the relative availability of NO_3^- to fringe trees (1 df contrast: $t = -3.43$, $P \leq 0.001$), but not dwarf and transition trees, a pattern inconsistent with the change in leaf $\delta^{15}N$ values.

Greater isotopic fractionation may also occur with slow plant growth because of low N demand (Goerick et al. 1994), and this pattern is consistent with the tree-height gradient. This plant fractionation model predicts that increased plant growth by addition of a growth-limiting nutrient other than N would increase N demand and decrease fractionation (because all N isotopes would be used). Enrichment with P, which enhanced growth and altered the nitrogen concentration of dwarf trees (Feller et al. 1999), was accompanied by an increase in leaf $\delta^{15}N$ values (Table 1, Fig. 2), consistent with model prediction. Greater resorption of P (73%) than N (47%) from senescing leaves indicates a higher demand for P by unfertilized dwarf trees (Feller et al. 1999). Furthermore, increased N use efficiency in response to P fertilization (61% increase, Feller et al. 1999) supports the explanation that N demand by *R. mangle* is increased by P addition. Finally, the strong

correlation between leaf $\delta^{15}N$ and leaf N:P is consistent with this model (Fig. 3). Those trees with highest demand for N (fringe controls and all P-fertilized trees) occur at the upper extreme of this range where N isotope discrimination is lowest, and trees with lowest demand (N-fertilized dwarf trees) fall at the lower extreme where discrimination is greatest. Plant fractionation, as a consequence of plant growth and N demand, thus appears to be a likely mechanism contributing to the different $\delta^{15}N$ values among height forms of *R. mangle* at Twin Cays. The dramatic elimination of this pattern by P addition, coupled with growth stimulation, supports the hypothesis that decreased tree height and productivity across the gradient involves P deficiency. Kao and Chang (1998) came to a similar conclusion regarding dwarf *Kandelia candel* (i.e., growth restriction was at least partly due to nutrient deficiency, based on low tissue concentrations of N and P compared to taller trees).

However growth of P-limited dwarf trees was more severely restricted than fringe or transition trees (Table 1). Since bioavailability of P did not vary across the tree-height gradient (Table 3, Fig. 1), P acquisition or assimilation may be limited secondarily by another factor or growth may be limited directly by other stresses that increase with distance landward. The negative correlation between leaf P concentrations (but not leaf N) and water depth at low tide suggests a connection between soil flushing and P delivery or acquisition (Fig. 5). Factors such as soil waterlogging and salinity vary along with growth and tree stature across the gradient (Tables 1 and 2; see also McKee 1993, 1995), and these relationships are considered further in the following discussion.

Carbon isotope composition

The increase in leaf $\delta^{13}C$ values from the fringe to dwarf zones paralleled the decrease in tree growth across the tree-height gradient (Fig. 2b, Table 2). This isotopic pattern may be due to decreased stomatal conductance, which resulted in lower internal carbon dioxide concentrations and lower carbon isotope discrimination (Farquhar et al. 1982, Lin and Sternberg 1992a, b); increased carboxylation efficiency via higher leaf N concentration and/or lower specific leaf area (leaf area per mass; Körner et al. 1988); or higher internal resistance to CO_2 diffusion due to thicker leaves (Vitousek et al. 1990, Lauteri et al. 1997).

The spatial pattern in C isotope discrimination was not significantly altered by nutrient addition (Fig. 2b). Some previous studies have also found no effect of fertilization on plant $\delta^{13}C$ (Lin and Sternberg 1992b, Michelsen et al. 1996), whereas others have (Guehl et al. 1995). Fertilization might cause opposing effects on the diffusive or enzymatic processes controlling C isotope discrimination and thus explain no change in $\delta^{13}C$ values. For example, fertilization of dwarf trees with P significantly reduces leaf thickness (Feller 1996), but

diffusive effects on $\delta^{13}C$ might be counterbalanced by an increase in carboxylation efficiency in response to increased availability of P. However the more likely explanation for lack of a fertilizer effect is that the processes affecting C isotope discrimination in *R. mangle* were not directly affected by nutrients. The consistent spatial pattern across control and N- and P-fertilized transects lends support to this interpretation (Fig. 2b).

Lin and Sternberg (1992*a*) examined carbon isotopic distributions in *R. mangle* in south Florida and found a pattern of increasing $\delta^{13}C$ signatures with decreasing tree height, similar to our study. Dwarf trees at Twin Cays ($-26.5‰$) had more negative $\delta^{13}C$ values than stunted trees (1 m tall) in Florida (from $-25‰$ to $-25.4‰$), whereas fringe *R. mangle* trees growing at sea-strength salinity at Twin Cays ($-28.4‰$) had a similar C isotope ratio to that in Florida (from $-28‰$ to $-29‰$). The difference between dwarf trees in Florida and Belize cannot be attributed to climatic factors, since fringe trees would have also differed. Another explanation is that salinity in the dwarf zone at Twin Cays is, on average, lower or fluctuates less than that in Florida. Lin and Sternberg (1992*b*) further found that growing *R. mangle* and other mangrove species at constant, sea-strength salinity decreased C isotope discrimination, which correlated with lower intercellular CO_2 concentrations and higher intrinsic transpiration efficiency. Plants grown in the greenhouse at 500 mmol/L NaCl (approximate concentration of seawater) had similar carbon isotope discrimination to fringe trees in the field, whereas that of plants grown at 100 mmol/L NaCl was ~3‰ higher. In another greenhouse study, Lin and Sternberg (1993) found that fluctuating salinity (biweekly or monthly) decreased growth of red mangrove seedlings more than a constant high salinity. However salinity fluctuations reduced growth only when varied from zero to 200 mol/m³ and from 100 to 400 mol/m³, but not from 250 to 750 mol/m³ (from ~18 to 53 g/L). The salinity range in dwarf stands at Twin Cays was from ~38 to 45 g/L (McKee 1993), which is not only narrow but within the experimental range in which Lin and Sternberg (1993) found no effect of fluctuating salinity on growth. In fact, salinity was highest and fluctuated most in the transition zone (McKee 1993), but tree height and isotopic discrimination were intermediate to that in the fringe and dwarf zones (Fig. 2).

Thus $\delta^{13}C$ values may increase in some situations as a consequence of elevated or fluctuating salinity that decreases stomatal conductance and intercellular CO_2 (Lin and Sternberg 1992*a, b,* 1993), but this factor alone cannot explain the pattern at Twin Cays. An alternative hypothesis is that the $\delta^{13}C$ pattern reflects spatial variation in some other factor influencing stomatal or carboxylation processes. In addition to salinity, stomatal conductance may also be decreased in mangroves by prolonged waterlogging, particularly in combination

with saline conditions (Naidoo 1985). Since the tree-height gradient was characterized by variation in inundation (Table 2), greater soil waterlogging may have altered stomatal conductance or photosynthetic rates, affecting C isotope discrimination. Carboxylation efficiency may also be increased by higher leaf tissue N concentrations or lower specific leaf area (Körner et al. 1988). Cordell et al. (1999) found that less negative $\delta^{13}C$ values were associated with increased carboxylation efficiency and leaf N on a per unit area basis in *Metrosideros polymorpha*. The significant correlation between mangrove $\delta^{13}C$ values and leaf N is consistent with this hypothesis (Fig. 3b). A final possibility is that leaf $\delta^{13}C$ values simply reflect variation in environmental conditions caused by changes in tree height and canopy development. For example, water temperature, which is affected by canopy openness and solar radiation, can vary across the tree-height gradient from 28°C in the fringe to >40°C in the dwarf zone (K. L. McKee and I. C. Feller, *personal observations*) and could have influenced leaf $\delta^{13}C$ values by decreasing stomatal conductance.

Conclusions

Although isotopic analyses cannot conclusively identify the factors contributing to growth variation in *R. mangle*, they suggest how nutrient switching may interact with other factors to limit growth. The results support two possible mechanisms that relate the known growth limiting factors at Twin Cays to explain observed patterns. Bioavailability of N relative to P increases with distance from shoreline, but growth is more restricted landward. There are two reasons for this conclusion.

(1) Delivery, uptake, or assimilation of P is more strongly affected by tidal flushing and concomitant factors that vary spatially than is N. The greater growth response to P enrichment (Table 1) and the correlation between leaf P concentration and tidal fluctuation (Fig. 5) support this hypothesis. As water movement and mass flow through the soil are decreased with distance from the shoreline, P delivery to roots may become more diffusionally restricted than N due to differences in mobility of ions. Strongly adsorbed ions such as PO_4^{-3} have diffusion coefficients as low as 10^{-9} cm²/s compared to NO_3^{-} and NH_4^{+} (10^{-5} and 10^{-7} cm²/s, respectively; Nye and Tinker 1977). Mangrove growth consequently diminishes with distance from the shoreline because the total amount of P delivered to roots decreases even though concentrations do not vary. Soil waterlogging can also restrict root growth or metabolism of mangroves (McKee 1996), and these changes may affect uptake or assimilation of P more than N across the gradient. Examination of flooding and salinity effects on P vs. N acquisition by mangroves may help to resolve this issue.

(2) Demand for P relative to N increases because stress effects differentially influence plant require-

ments for these nutrients. The alteration of N isotope fractionation by P addition to slow-growing plants and the correlation between leaf $\delta^{15}N$ values and leaf N:P concentrations support the argument that the patterns reflect external nutrient availability relative to internal demand. Increased stress across the gradient may have increased the plant requirement for P relative to N. Leaf $\delta^{13}C$ values indicated spatial variation in stress factors (independent of nutrient limitations) affecting stomatal conductance or carboxylation (Fig. 2), even though the analysis could not pinpoint which ones were involved in growth restriction. The effects of nutrient availability, flooding, and salinity need to be examined under controlled conditions and/or in conjunction with physiological measurements to fully interpret the effects of these stresses on mangrove growth, nutrient relations, and isotope discrimination.

This work aids in understanding the N to P limitation to mangrove growth across a tree-height gradient in Belize as well as environmental factors influencing mangrove discrimination of carbon and nitrogen isotopes. Our findings hint at the ecological complexity of these floristically simple ecosystems and illustrate the value of isotopic analysis in examining nutrient dynamics and stress responses by mangroves and other vegetation. The results also have implications for identifying nutrient loading in coastal systems dominated by mangroves. Fertilization with N and P clearly generates different isotopic compositions in mangrove tissues, but the pattern will depend on supplies of nutrients relative to internal demand and other environmental factors influencing nutrient acquisition. Additional work is needed to further refine these relationships, to identify those conditions that can alter isotopic distributions, and to clarify the plant processes controlling fractionation of N and C.

Literature Cited

Ball, M. C. 1988. Ecophysiology of mangroves. Trees **2**:129–142.

Boto, K. G., and J. T. Wellington. 1983. Phosphorus and nitrogen nutritional status of a northern Australian mangrove forest. Marine Ecology Progress Series **11**:63–69.

Boto, K. G., and J. T. Wellington. 1984. Soil characteristics and nutrient status in a northern Australian mangrove forest. Estuaries **7**:61–69.

Bradley, P. M., and J. T. Morris. 1990. Influence of oxygen and sulfide concentration on nitrogen uptake kinetics in *Spartina alterniflora*. Ecology **71**:282–287.

Carpenter, E. J., R. R. Rahvey, B. Fry, and D. G. Capone. 1997. Biogeochemical tracers of the marine cyanobacterium *Trichodesmium*. Deep-sea Research **44**:27–38.

Cordell, S., G. Goldstein, G. F. C. Meinzer, and L. L. Handley. 1999. Allocation of nitrogen and carbon in leaves of *Metrosideros polymorpha* regulates carboxylation capacity and $\delta^{13}C$ along an altitudinal gradient. Functional Ecology **13**:811–818.

Ellison, A. M., E. J. Farnsworth, and R. R. Twilley. 1996. Facultative mutualism between red mangroves and root-fouling sponges in Belizean mangal. Ecology **77**:2431–2444.

Evans, R. D., A. J. Bloom, S. S. Sukrapanna, and J. R. Ehleringer. 1996. Nitrogen isotope composition of tomato (*Lycopersicon esculentum* Mill cv. T-5) grown under ammonium or nitrate nutrition. Plant, Cell and Environment **19**:1317–1323.

Farquhar, G. D. 1993. Carbon and oxygen isotope effects in the exchange of carbon dioxide between terrestrial plants and the atmosphere. Pages 47–70 *in* J. R. Ehleringer, A. E. Hall, and G. D. Farquhar, editors. Stable isotopes and plant carbon-water relations. Academic Press, San Diego, California, USA.

Farquhar, G. D., M. C. Ball, S. von Caemmerer, and Z. Roksandic. 1982. Effect of salinity and humidity on $\delta^{13}C$ values of halophytes–evidence for diffusional isotope fractionation determined by the ratios of intercellular/atmospheric CO_2 under different environmental conditions. Oecologia **52**:121–137.

Feller, I. C. 1995. Effects of nutrient enrichment on growth and herbivory of dwarf red mangrove (*Rhizophora mangle*). Ecological Monographs **65**:477–505.

Feller, I. C. 1996. Effects of nutrient enrichment on leaf anatomy in red mangrove. Biotropica **28**:13–22.

Feller, I. C., K. L. McKee, D. F. Whigham, and J. P. O'Neill. 2001. Nitrogen vs. phosphorus limitation across an ecotonal gradient in a mangrove forest. Biogeochemistry, In press.

Feller, I. C., D. F. Whigham, J. P. O'Neill, and K. L. McKee. 1999. Effects of enrichment on within-stand nutrient cycling in a red-mangrove (*Rhizophora mangle* L.) forest. Ecology **80**:2193–2205.

Fry, B., A. L. Bern, M. S. Ross, and J. F. Meeder. 2000. $\delta^{15}N$ studies of nitrogen use by the red mangrove, *Rhizophora mangle* L., in South Florida. Estuarine, Coastal and Shelf Science **50**:723–735.

Goerick, R., J. P. Montoya, and B. Fry. 1994. Physiology of isotopic fractionation in algae and cyanobacteria. Pages 187–221 *in* K. Lajtha and R. Michener, editors. Stable isotopes in ecology and environmental science. Blackwell Science, Oxford, UK.

Guehl, J., C. Fort, and A. Ferhi. 1995. Differential response of leaf conductance, carbon isotope discrimination and water-use efficiency to nitrogen deficiency in maritime pine and pedunculate oak plants. New Phytologist **131**:149–157.

Handley, L. L., and C. M. Scrimgeour. 1997. Terrestrial plant ecology and ^{15}N abundance: the present limits to interpretation for uncultivated systems with original data from a Scottish old field. Advances in Ecological Research **27**:133–212.

Jordan, M. J., K. J. Nadelhoffer, and B. Fry. 1997. Nitrogen cycling in forest and grass ecosystems irrigated with treated wastewater enriched in ^{15}N. Ecological Applications **7**:864–881.

Kao, W.-Y., and K.-W. Chang. 1998. Stable carbon isotope ratio and nutrient contents of the *Kandelia candel* mangrove populations of different growth forms. Botanical Bulletin of Academia Sinica **39**:39–45.

Koch, M., I. A. Mendelssohn, and K. L. McKee. 1990. Mechanism for the hydrogen sulfide-induced growth limitation

in wetland macrophytes. Limnology and Oceanography **35**: 399–408.

Koltes, K. H., J. J. Tschirky, and I. C. Feller. 1998. Carrie Bow Cay, Belize. Pages 79–94 *in* B. Kjerfve, editor. CARICOMP—Caribbean coral reef, seagrass, and mangrove sites. Coastal region and small island papers 3, United Nations Educational, Scientific and Cultural Organization (UNESCO), Paris, France.

Körner, C., G. D. Farquhar, and Z. Roksandic. 1988. A global survey of carbon isotope discrimination in plants from high altitude. Oecologia **74**:623–632.

Lajtha, K. 1988. The use of ion-exchange resin bags for measuring nutrient availability in an arid ecosystem. Plant Soil **105**:105–111.

Lauteri, M., A. Scatazza, M. C. Guido, and E. Brugnoli. 1997. Genetic variation in photosynthetic capacity, carbon isotope discrimination and mesophyll conductance in provenances of *Castanea sativa* adapted to different environments. Functional Ecology **11**:675–683.

Lin, G., and L. da S. L. Sternberg. 1992*a*. Differences in morphology, carbon isotope ratios, and photosynthesis between scrub and fringe mangroves in Florida, USA. Aquatic Botany **42**:303–313.

Lin, G., and L. da S. L. Sternberg. 1992*b*. Effect of growth form, salinity, nutrient and sulfide on photosynthesis, carbon isotope discrimination and growth of red mangrove (*Rhizophora mangle* L.). Australian Journal of Plant Physiology **19**:509–517.

Lin, G., and L. da S. L. Sternberg. 1993. Effect of salinity fluctuation on photosynthetic gas exchange and plant growth of the red mangrove (*Rhizophora mangle* L.). Journal of Experimental Botany **44**:9–16.

Lugo, A. E., and S. C. Snedaker. 1974. The ecology of mangroves. Annual Review of Ecology and Systematics **5**:39–64.

Macko, S. A., and N. E. Ostrom. 1994. Pollution studies using stable isotopes. Pages 45–62 *in* K. Lajtha and R. Michener, editors. Stable isotopes in ecology and environmental science. Blackwell Science, Oxford, UK.

McKee, K. L. 1993. Determinants of mangrove species distribution in neotropical forests: biotic and abiotic factors affecting seedling survival and growth. Dissertation. Louisiana State University, Baton Rouge, Louisiana, USA.

McKee, K. L. 1995. Seedling recruitment patterns in a Belizean mangrove forest: effects of establishment ability and physico-chemical factors. Oecologia **101**:448–460.

McKee, K. L. 1996. Growth and physiological responses of mangrove seedlings to root zone anoxia. Tree Physiology **16**:883–889.

McKee, K. L., I. A. Mendelssohn, and M. W. Hester. 1988. Reexamination of pore water sulfide concentrations and redox potentials near the aerial roots of *Rhizophora mangle* and *Avicennia germinans*. American Journal of Botany **75**: 1352–1359.

Mendelssohn, I. A. 1979. Nitrogen metabolism in the height forms of *Spartina alterniflora* in North Carolina. Ecology **60**:574–584.

Michelsen, A., S. Jonasson, D. Sleep, M. Havström, and T. V. Callaghan. 1996. Shoot biomass, $\delta^{13}C$, nitrogen and chlorophyll responses of two arctic dwarf shrubs to in situ shading, nutrient application and warming simulating climatic change. Oecologia **105**:1–12.

Montoya, J. P., and J. J. McCarthy. 1995. Isotopic fractionation during nitrate uptake by phytoplankton grown in continuous culture. Journal of Plankton Research **17**:439–464.

Morris, J. T. 1984. Effects of oxygen and salinity on ammonium uptake by *Spartina alterniflora* Loisel. and *Spartina patens* (Aiton) Muhl. Journal of Experimental Marine Biology and Ecology **78**:87–98.

Nadelhoffer, K., and B. Fry. 1994. Nitrogen isotope studies in forest ecosystems. Pages 22–44 *in* K. Lajtha and R. Michener, editors. Stable isotopes in ecology and environmental science. Blackwell Science, Oxford, UK.

Nadelhoffer, K., G. Shaver, B. Fry, A. Giblin, L. Johnson, and R. McKane. 1996. ^{15}N natural abundances and N use by tundra plants. Oecologia **107**:386–394.

Naidoo, G. 1985. Effects of waterlogging and salinity on plant–water relations and on the accumulation of solutes in three mangrove species. Aquatic Botany **22**:133–143.

Nye, P. H., and P. B. Tinker. 1977. Solute movement in the soil–root system. Blackwell Science, Oxford, UK.

Pennock, J. R., D. J. Velinsky, J. M. Ludlam, J. H. Sharp, and M. L. Fogel. 1996. Isotopic fractionation of ammonium and nitrate during uptake by *Skeletonema costatum*: implications for $\delta^{15}N$ dynamics under bloom conditions. Limnology and Oceanography **41**:451–459.

Potts, M. 1979. Nitrogen fixation (acetylene reduction) associated with communities of heterocystous and non-heterocystous blue-green algae in mangrove forests of Sinai. Oecologia **39**:359–373.

Primavera, J. H. 1998. Mangroves as nurseries: shrimp populations in mangrove and non-mangrove habitats. Estuarine, Coastal and Shelf Science **46**:457–464.

Rolston, D. E., G. E. Fogg, D. L. Decker, D. T. Louie, and M. E. Grismer. 1996. Nitrogen isotope ratios identify nitrate contamination sources. California Agriculture **50**:32–36.

Turner, R. E. 1977. Intertidal vegetation and commercial yields of penaeid shrimp. American Fisheries Society Transactions **106**:411–416.

Vitousek, P. M. 1999. Nutrient limitation to nitrogen fixation in young volcanic sites. Ecosystems **2**:505–510.

Vitousek, P. M., C. B. Field, and P. A. Matson. 1990. Variation in foliar abundance in Hawaiian *Metrosideros polymorpha*: a case of internal resistance? Oecologia **84**:362–370.

Yoneyama, T. 1995. Nitrogen metabolism and fractionation of nitrogen isotopes in plants. Pages 92–102 *in* K. Lajtha and R. Michener, editors. Stable isotopes in ecology and environmental science. Blackwell Science, Oxford, UK.

Yoneyama, T., T. Omata, S. Nakata, and J. Yazaki. 1991. Fractionation of nitrogen isotopes during the uptake and assimilation of ammonia by plants. Plant and Cell Physiology **32**:1211–1217.