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Isotopic signatures and nutrient relations of plants inhabiting brackish wetlands in the northeastern coastal plain of Venezuela

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Abstract The semi-diurnal tidal regime (≥ 2 m) in the Paria Gulf on the Atlantic coast of Venezuela, and the flat landscape of the region, allow the penetration for tens of km of marine waters into the rivers draining the northeastern coastal plain of the country. The levels of salinity, tidal flooding, and sedimentation decrease perpendicularly from the river channel toward the back swamps. The vegetation varies sequentially from fringe mangroves along the river margins, to back swamps containing forests dominated by *Pterocarpus officinalis*, herbaceous communities of *Lagenocarpus guianensis*, and palm swamps with *Mauritia flexuosa*, *Chrysobalanus icaco*, and *Tabebuia* spp. This environmental structure was used to test the hypotheses that: (a) mangrove distribution is strongly associated with salinity of interstitial water, and (b) they occupy areas where tidal influence and sediment dynamics determine a relatively open N cycle. Analyses of soil, water, and plants along a 1.5 km transect located near the confluence of the Guanoco and San Juan Rivers (Sucre and Monagas States, Venezuela) revealed that: (a) conductivity decreased from 11 to 0.2 mmhos cm^{-1} from the river fringe to the internal swamp, whereas Na in the same stretch decreased from 100 to 2 μM; (b) average leaf tissue concentrations of Na, P, and N decreased significantly along the transect; (c) *P. officinalis* showed a large Na-exclusion capacity indicated by positive K/Na ratios from 8 to 200, and *Crinum erubescens* counteracted Na by accumulating K above 1,000 mmol kg^{-1}; (d) leaves varied widely in δ ^{13}C (−25.5 to −32‰) and δ ^{15}N (4 to −10.5‰) values. Samples were aggregated according to soil carbon content corresponding to those of the mangrove forest belt (5–28 mol C kg^{-1}; 0–650 from river fringe) and those of the back swamps (40–44 mol C kg^{-1}; 700–1,500 m from river fringe). The concentrations of Na, P, and N (in mmol kg^{-1}) and δ ^{15}N values (in ‰) were significantly higher in the mangrove forest compared to the back swamp (Na 213 vs. 88; P 41 vs. 16; N 1,535 vs. 727; δ ^{15}N 1.5 vs. −3.7), indicating that the fringe forest was not nutrient limited. These results support the hypotheses that mangroves are restricted to the more-saline sections of the transect, and that the fringe forest has a more open N cycle, favoring ^{15}N accumulation within the system.

Keywords Coastal wetlands · Mangroves · Nutrients · Salinity · δ ^{13}C · δ ^{15}N

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Introduction

On the northeastern coast of South America, north of the Orinoco Delta, there is a swampy coastal plain that developed during the Holocene due to the deposition of sediments of fluvio-marine origin (COPLANARH 1974). This region is characterized by a macro tidalregime (≥ 2 m) that, given the flat landscape with slopes of 0–0.5%, allows the penetration of seawater upstream in the rivers that drain the area. Annual rainfall is highly seasonal, averaging 1,800–2,100 mm. River levees and associated fringing mangrove forests are subjected to tidal flooding twice daily, while the back swamps are flooded permanently with a water depth that varies seasonally according to the rainfall distribution (COPLANARH 1974). These hydrologic and geomorphologic conditions create a complex pattern of salinity and flooding gradients, clearly identified along perpendicular transects from the river channels by changes in vegetation density and composition (Medina and Francisco 1997; Colonnello and Medina 1998). The heterogeneous landscape may be divided into two main geomorphological units (COPLANARH 1974; MARNR 1979, 1990):

(a) Back swamps, where the landscape is flooded by spring tides and/or rainwater. Flooding and subsidence determine the development of organic soils and abundant peat (tropofibrists, tropohemists), where the vegetation is usually herbaceous or forested swamps;
(b) River banks along drainage channels, flooded daily by tides, with high-organic pyrite-rich clay soils (sulfic hydraquents) and covered by a dense mangrove forest.

These distinct geomorphological units offer an opportunity to investigate the correlations between salinity and hydroperiod with vegetation zonation and the nutritional profiles of the dominant species within each vegetation type. The ecophysiological constraints leading to the formation of well-defined vegetation zones include salinity, along the river fringes because of the penetration of salt water, semidiurnal tides, and the seasonal rainfall pattern. These constraints regulate the coexistence of species with different degrees of tolerance to salt and flooding (low oxygen partial pressure around the root environment). We hypothesize that:

(1) Back swamps have a relatively closed nutrient cycle because of their relative isolation from the drainage channels, and are nutrient limited because of slow rates of organic matter decomposition, and low input of river water;
(2) Mangrove forests on river levees grow on saline soils, and have a more-open nutrient cycle determined by sediments transported in and out by the tides.

Therefore, we expect to find significant changes in nutrient concentrations, particularly N and P, in plant tissues. In addition, the natural abundance of the ^{15}N isotope is expected to increase with N availability and openness of the nutrient cycle (Martinelli et al. 1999), whereas that of the ^{13}C isotope may reveal the effect of salinity on leaf conductance to water vapor (Ball and Farquhar 1984).

This paper builds upon a previous study on the structure of mangrove forest and associated plant communities in one sector of the Venezuelan northeastern coastal plain (Fig. 1) (Canales et al. in MARNR 1990), and a vegetation study in the vegetation of the Guanoco wetlands by the Venezuelan oil company (Petróleos de Venezuela S.A. PDVSA) (González et al. 1999).

Study area and methods

The study site is located in the southern margin, near the mouth of the Guanoco River into the San Juan River (Lat 10°05′, Long 62°52′30″, Fig. 1). In this location PDVSA built a narrow, 700-m-long railway perpendicular to the fringe zone of the Guanoco River through a herbaceous swamp located behind the mangrove belt, that allowed access into a swamp forests further inland (Fig. 1). We used this railway to collect soils, interstitial water, and leaves of the dominant species, within sites selected following the vegetation description of González et al. (1999).

We sampled soils from 0–30 cm, encompassing the effective root penetration in saturated soils, interstitial water below 10 cm soil depth, which was assumed to represent the soil solution around active roots, and the leaves of dominant species at nine sites located at increasing distances from the river fringe (0–1,500 m). The sequence of sampling points and species sampled is described in Table 1.

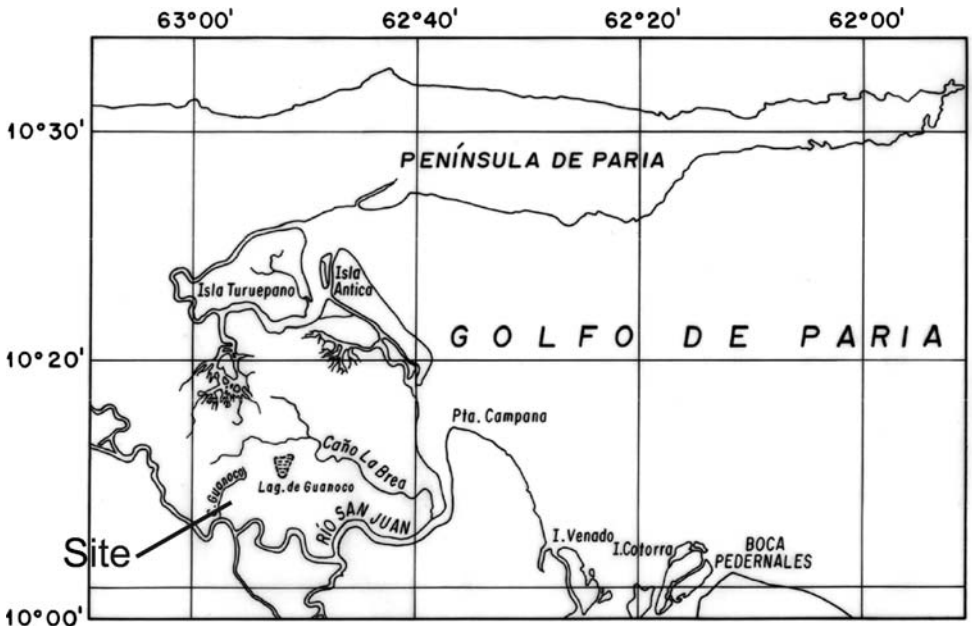

Fig. 1 Location of the study area

The specific conductivity of interstitial water samples was measured with a conductivity meter using a Pt dipcell (Cole-Parmer EC Meter 19101-00), values are expressed as mmhos cm^{-1}. The cation concentration of interstitial water was measured at PDVSA using a plasma spectrometer calibrated against inorganic standard solutions.

Soil samples were highly organic and water-saturated in the field, therefore they were collected in plastic bags and brought to the laboratory within 24 h of collection. In the laboratory, root material, coarse woody debris, and partially decomposed leaves were removed by hand. The remainder of the sample was then dried in a ventilated oven at 60°C to constant weight, and ground in a porcelain mortar to pass a 1 mm sieve. Leaf samples were carefully cleaned with tissue paper before drying in a ventilated oven at 60°C to constant weight. Dry leaves were finely ground in a modified coffee-bean grinder until particles were barely visible. Leaf and soil samples were analyzed at the International Institute of Tropical Forestry, USDA-Forest Service, Puerto Rico. Aliquots of the homogenized samples were digested in a mixture of nitric acid-hydrogen peroxide and analyzed for P, Na, K, Mg, and Ca using ICP (Induced Current Plasma) techniques (Luh Huang and Schulte 1985). Briefly, the procedure consisted of weighing a 50–100 mg sample in a 100 ml glass tube. To each tube 5 ml concentrated HNO_3 was added and left to stand overnight. The tube was then heated at about 60°C for half an hour. After cooling 3 ml 30% H_2O_2 was added and the mixture was heated to about 165°C for 1.5 h. After cooling 2.5 ml LiCl was added, made up to 100 ml with distilled water filtered through ash-free filter paper. Digests were analyzed with a Beckman DCP-AES model Spectraspan V.

We report the element concentrations obtained from the plasma spectrometer calibrated against inorganic standards. Recovery factors for Na, K, Mg, Ca, and P were obtained by running organic standards (Pine 1575a, National Institute of Standards and Technology); their values averaged 100%, 81%, 85%, 81%, and 74%, respectively. Carbon, nitrogen, and sulfur in leaves and soil samples were measured with a combustion technique in a LECO CNS (Carbon Nitrogen Sulfur) analyzer (Tabatai and Bremmer 1991). The recovery factor of this method estimated by running National Institute of Standards and Technology (NIST) plant standards was >99%. Percentage ash was estimated by loss on ignition (LOI) when heating the sample (0.5 g) in a muffle at 450°C for 8 h. Samples for isotopic analyses were ground as indicated above. Acidification treatment of

Table 1 Sampling sites and characteristic species

Distance from river (m)	Description	Species sampled
1. 0–10 m	Gallery forest belt lining the river (lower canopy samples)	*Rhizophora racemosa* G. Mey.
		Avicennia germinans L.
2. 50 m	Belt of *R. racemosa*, with *Crinum erubescens* dominating the forest floor (lower canopy samples)	*R. racemosa*
		Pterocarpus officinalis Jacq.
		Crinum erubescens Ayton
3. 300 m	*Rhizophora-Pterocarpus* belt (lower canopy samples)	*R. racemosa*
		P. officinalis
4. 500 m	*Pterocarpus-Rhizophora* belt (lower canopy samples)	*P. officinalis*
		R. racemosa
5. 650 m	*Laguncularia* belt with isolated *Pterocarpus* individuals (full exposure samples)	*Laguncularia racemosa* (L.) Gaert. f.
		Acrostichum aureum L.
		Acrostichum danaefolium Lang.&Fish.
		Montrichardia arborescens (L.) Shott
		R. racemosa
6. 750 m	Dense *Lagenocarpus* meadow, >2 m height, with isolated individuals of *A. aureum* and *A. danaefolium*	*Lagenocarpus guianensis* Lindl&Nees
7. 850 m	Shrubby community of *Chrysobalanus* with *Blechnum*	*Blechnum serrulatum* L.C. Richard
		Tabebuia insignis var. *monophylla Sandw.*
		Chrysobalanus icaco L.
		Clusia sp. 1
		Clusia sp. 2
		Fuirena umbellata Rottb.
8. 1,250 m	Dense shrubby community of *Chrysobalanus* and *Tabebuia*, with *Blechnum* and *Aechmea aquilega*	*Clusia* sp. 2
		B. serrulatum
		C. icaco
9. 1,500 m	Herbaceous swamp of *Lagenocarpus*, <1 m height, with isolated individuals of *Tabebuia* and *Blechnum*	*L. guianensis*
		T. insignis var. *monophylla*
		B. serrulatum
		Mauritia flexuosa L.
	Swamp forest with *Mauritia*, *Chrysobalanus*, *Clusia* sp. and other unidentified tree species	*Clusia* sp. 1
		C. icaco

soils samples was not necessary because there was no calcium carbonate present. Carbon and nitrogen isotope ratios (δ ^{13}C and δ ^{15}N) were measured at the SERC Stable Isotope Laboratory (Florida International University, Florida USA) using standard elemental analyzer isotope-ratio mass spectrometer (EA-IRMS) procedures. The EA is used to combust the organic material and to form N_2 and CO_2, which were measured on a Finnigan MAT delta C IRMS in a continuous flow mode using ultra pure He as a carrier gas. The samples' isotopic ratios (R) are reported in the standard delta notation (‰): δ (‰) = $[(R_{sample}/R_{standard}) - 1] \times 1{,}000$. These stable isotopic results are reported with respect to the international standards of atmospheric nitrogen (AIR, N_2) and Vienna Pee Dee belemnite (V-PDB) for

carbon. The analytical reproducibility of this study based on replicates of internal standards was better than ±0.2‰ for δ ^{15}N, and ±0.08‰ for δ ^{13}C.

Concentration of cations, N, P, S, and isotope ratios were regressed against distance from the river fringe, as a surrogate for environmental gradient, and the determination coefficient of the linear regression (r^2 = explained variance/total variance, Gotelli and Ellison 2004, p. 249) was used to assess differences in concentration patterns. This coefficient was adjusted to account for the number of samples (n).

Regression analysis was also used to assess the correlation between ion concentration and conductivity in water samples, percentage ash, and $\sum$cation, and the N and P concentrations in leaf tissues. A one-way analysis of variance was used to analyze the differences in concentration between the species of forest belt on the natural levees, and the herbaceous and forest species on the back swamps. This comparison showed that several parameters had inequal variances (Bartlett test), in those cases an analysis of variance allowing for different standard deviations, the Welch's test, was applied. All statistical procedures were conducted with a statistical software package (JMP, SAS Institute Inc. 2002).

Results

Sequence of vegetation units along the transect

The vegetation units recognized in this paper coincide with those reported for the same 750 m profile by González et al. (1999), and are succinctly described in Table 1. The river fringe was occupied by a mangrove forest dominated by *Avicennia germinans* and *Rhizophora racemosa*. All samples of *Rhizophora* that we collected along the transect were identified as *R. racemosa* according to the description of Tomlinson (1986). We did not find the species *R. harrisonii* reported by González et al. (1999). *R. racemosa* dominates behind the mangrove fringe, forming a belt approximately 300 m wide. Farther inland, the legume tree *P. officinalis* became more frequent, reaching dominance 300–500 m from the river fringe. Beyond the 500 m line, the forest canopy became much lower, forming a dense thicket dominated by *Laguncularia racemosa* with isolated individuals of *P. officinalis*. The presence of this mangrove species was unexpected because it is a halophyte, and we assumed that reduction of salinity from the river fringe landwards would favor the development of more-flood-tolerant, but salt-sensitive woody plants. Beginning at the 700 m line a tall, dense, herbaceous swamp dominated by the sedge *Lagenocarpus guianensis* with isolated individuals of the ferns *Acrostichum aureum* and *A. danaefolium* (distinguished in the field by the fraction of fertile leaflets in the reproductive frond, Tomlinson 1986) appeared. Beyond the 800 m line a low, mixed shrubland of variable density was found, dominated by shrubby individuals of *Chrysobalanus icaco*, *Tabebuia insignis*, and two unidentified *Clusia* species. The floor of this community was densely covered by the sedge *Fuirena umbellata* and the fern *Blechnum serrulatum*. Interspersed within the shrubby belt were several herbaceous swamps with *L. guianensis*, *B. serrulatum*, isolated shrubs of *T. insignis* and the palm tree *Mauritia flexuosa*.

Soil organic matter and ionic concentration of interstitial water

The upper soil layer (0–30 cm) showed high carbon content along the whole transect, varying from 5 to 18 mol kg^{-1} in the first 650 m (Fig. 2A). From the herbaceous swamp onwards, the carbon concentration of the substrate increased to 40–45 mol C kg^{-1}. Nitrogen showed a similar distribution pattern as C (Fig. 2A), but the N/C ratio, corresponding to the concentration of N in soil organic matter, decreased in the same direction. Sulfur increased from the river fringe up to the outer mangrove belt, decreasing steadily afterwards (Fig. 2B). The phosphorus level was similar within the mangrove belt sites and decreased rapidly towards the back swamp (Fig. 2B).

Specific conductivity of interstitial water decreased nearly exponentially from the river fringe to the herbaceous and forest swamps (Fig. 3). Site 5 (650 m from river fringe) did not follow this trend, with a conductivity value similar to the previous mangrove dominated vegetation. The concentration of Na^+, K^+, Mg^{2+}, and Ca^{2+} followed the same pattern along the transect, with a clear increase in concentration at site 5, Na always being the most abundant cation (Fig. 3). Calcium was more concentrated at sites 2, 5, and 6. The relative reduction

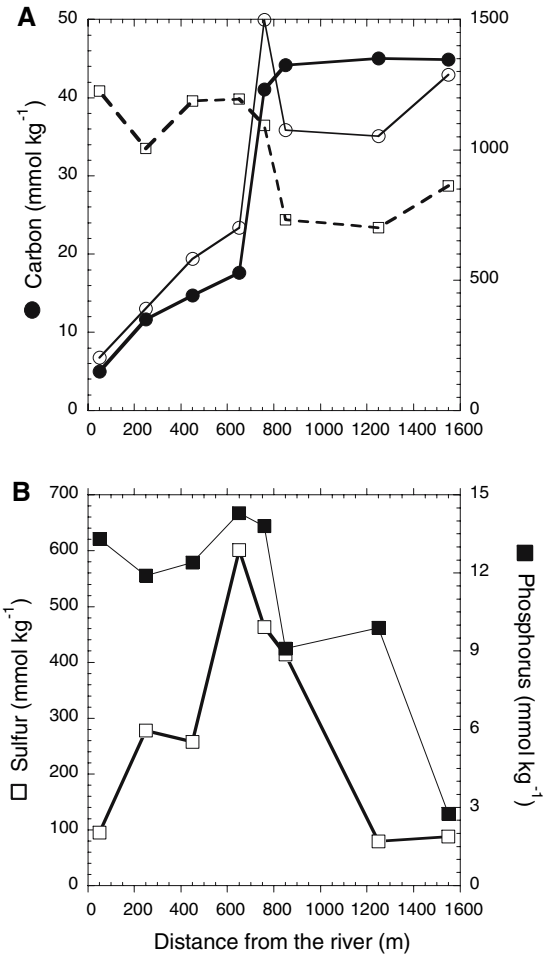

Fig. 2 Concentration of C, N, S, and P of soils along the 1,500 m transect on the southern side of the Guanoco River

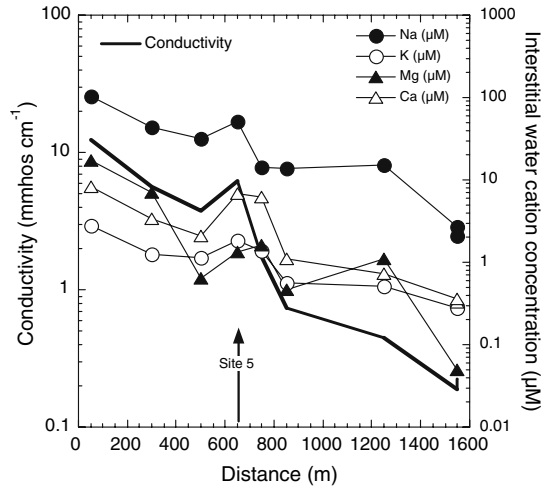

Fig. 3 Conductivity and cation concentration of interstitial water along the 1,500 m transect on the southern side of the Guanoco River

factors of ion concentration between sites 2 and 9 were 10 for K, 25 for Ca, 49 for Na, and 344 for Mg. Interstitial water conductivity was linearly related to the concentration of each cation, but the determination indices (r^2) decreased in the sequence Na (0.96) > Mg (0.85) > K (0.80) > Ca (0.63).

Cation concentration in leaf tissues

The total concentration of cations ($\sum$[Na, K, Mg, Ca]) showed a large range of variation in the sampled species (Table 2). One group of species was characterized by cation accumulations below 600 mmol kg^{-1} (*M. flexuosa*, *L. guianensis*, *F. umbellata*, and *C. icaco*); at the other extreme, a species group with accumulations above 1,000 mmol kg^{-1} included the three mangrove species (*R. racemosa*, *L. racemosa*, and *A. germinans*) and a herbaceous species that covers the forest floor of the coastal mangrove belt submitted to shallow daily flooding (*C. erubescens*).

The sum of cations was linearly correlated with the percentage ash content, with three notable exceptions: *A. danaefolium* at site 5, and *C. icaco* and *F. umbellata* at site 7. Excluding these species, the regression explained more than 60% of the percentage ash variation, and the average amount of ash material not related to the main cations amounted to 2.1% (ash% = 2.125 + 0.006 $\sum$Cation mmol kg^{-1}; $r^2_{adjusted}$ = 0.601; $P < 0.001$). The dispersion of percentage ash values in relation to the total cation concentration was probably associated with interspecific differences in accumulation of silica, but this element was not measured.

The concentration of individual cations did not show a specific tendency along the transect, with the exception of Na (Table 2). The sodium concentration decreased progressively from the river fringe to the forest and herbaceous swamps. *Pterocarpus officinalis* departed from this pattern, showing a substantial exclusion of Na in spite of rooting within the same soil environment occupied by mangrove species. The species *C. icaco* and *M. flexuosa* also showed a high Na-exclusion capacity compared with other species growing within the same habitat.

Table 2 Ash and cation concentration of leaf tissues along the Guanoco transect

Distance (m)	Species	Ash %	K (mmol kg^{-1} dry weight)	Na (mmol kg^{-1} dry weight)	Mg (mmol kg^{-1} dry weight)	Ca (mmol kg^{-1} dry weight)	∑Cat. (mmol kg^{-1} dry weight)	K/Na	Ca/Mg
0	*Avicennia germinans*	9.9	284	662	263	131	1,339	0.4	0.5
	Rhizophora racemosa	7.2	53	340	106	279	777	0.2	2.6
50	*Crinum erubescens*	14.0	1,008	210	222	309	1,749	4.8	1.4
	Pterocarpus officinalis	4.5	336	5	113	222	676	71.6	2.0
	Rhizophora racemosa	7.7	159	305	102	335	900	0.5	3.3
300	*Pterocarpus officinalis*	4.9	331	5	116	164	615	70.4	1.4
	Rhizophora racemosa	9.5	134	417	119	444	1,114	0.3	3.7
500	*Pterocarpus officinalis*	5.2	368	2	149	187	705	204.2	1.3
	Rhizophora racemosa	7.3	145	276	87	314	822	0.5	3.6
650	*Acrostichum aureum*	12.1	321	104	100	100	624	3.1	1.0
	Acrostichum danaefolium	7.4	614	89	82	62	847	6.9	0.8
	Laguncularia racemosa	8	374	153	127	391	1,046	2.5	3.1
	Montrichardia arborescens	8.6	361	282	111	192	946	1.3	1.7
	Rhizophora racemosa	8.4	103	136	110	541	890	0.8	4.9
750	*Lagenocarpus guianensis*	4.1	196	177	42	41	456	1.1	1.0
850	*Blechnum serrulatum*	6.4	314	91	89	99	591	3.5	1.1
	Chrysobalanus icaco	9.5	176	1	27	236	439	195.0	8.9
	Clusia sp. 1	4.9	89	71	86	316	562	1.3	3.7
	Clusia sp. 2	7.8	185	69	121	461	836	2.7	3.8
	Fuirena umbellata	9.8	130	223	113	63	529	0.6	0.6
	Tabebuia insignis	6.6	132	138	169	227	666	1.0	1.4
1,250	*Blechnum serrulatum*	8.2	235	130	107	128	599	1.8	1.2
	Chrysobalanus icaco	7	240	17	67	277	600	13.9	4.1
	Clusia sp. 2	6.9	188	92	183	482	945	2.0	2.6
	Tabebuia insignis	5.3	219	251	131	139	740	0.9	1.1

Table 2 continued

Distance (m)	Species	Ash %	K (mmol kg^{-1} dry weight)	Na (mmol kg^{-1} dry weight)	Mg (mmol kg^{-1} dry weight)	Ca (mmol kg^{-1} dry weight)	∑Cat. (mmol kg^{-1} dry weight)	K/Na	Ca/Mg
1,500	*Blechnum serrulatum*	5.4	368	47	71	89	575	7.9	1.3
	Chrysobalanus icaco	6.3	189	13	81	303	586	15.0	3.7
	Clusia sp. 1	6.2	153	68	55	517	793	2.3	9.5
	Lagenocarpus guianensis	4.1	198	30	68	44	339	6.6	0.6
	Mauritia flexuosa	2.9	150	1	36	32	218	136.2	0.9
	Tabebuia insignis	5.5	239	74	144	288	745	3.2	2.0
	Average	7.1	258	144	110	239	751	24.6	2.5

Average cation concentration of all plant samples followed the sequence K > Ca >> Na > Mg with 258, 239, 144, and 110 mmol kg^{-1}, respectively. *P. officinalis*, the two *Acrostichum* species, *L. racemosa*, *M. arborescens*, and particularly *C. erubescens* had above average K concentrations, while the mangrove *R. racemosa*, *C. icaco*, *T. insignis* and the *Clusia* species always had below-average K concentrations. Both *Clusia* species, and the mangrove species *L. racemosa* and *R. racemosa* behaved as relative Ca accumulators, whereas the sedges *M. flexuosa* and the ferns showed Ca concentrations well below average. Sodium concentrations were above 100 mmol kg^{-1} in all the mangrove species, *C. erubescens*, and unexpectedly in the sedge *F. umbellata* and the tree *T. insignis*. The lowest values, usually below 20 mmol kg^{-1}, were recorded in *P. officinalis*, *M. flexuosa* and *C. icaco*. In the case of Mg, *C. icaco*, *L. guianensis*, and *M. flexuosa* showed values below 60 mmol kg^{-1}, whereas *T. insignis*, *Clusia* sp. 2, *C. erubescens*, and particularly the mangrove *A. germinans*, showed the highest concentrations (up to 250 mmol kg^{-1}).

The K/Na ratios allowed the separation of three species groups in the transect (Table 2): (a) ratios between 1 and 10, including 17 out of the 32 plant samples; (b) ratios from 10 to 200, including the species *C. icaco*, *M. flexuosa*, and *P. officinalis*; (c) ratios below 1, including two mangrove species (*R. racemosa* and *A. germinans*), *F. umbellata*, and *T. insignis*. The latter two species occurred beyond site 6, with the lowest Na concentration in interstitial water. This result suggests that these species have low efficiency of K accumulation, rendering them sensitive to salinization.

The Ca/Mg ratios separated two extreme groups of species, one with ratios > 2 that included all the mangroves species except *A. germinans*, and one with ratios ≤ 1, including both sedges, both *Acrostichum* species, *M. flexuosa*, and the mangrove *A. germinans*.

Leaf tissue concentration of N, S, and P

The concentration of these elements associated to proteins and nucleic acids varied markedly among species because of differences in ash and organic components not associated with cytoplasm such as fiber (Table 3). Nitrogen concentration was below 600 mmol kg^{-1} in both species of *Clusia*, and in the sedge *L. guianensis*, but it was above 1,500 mmol kg^{-1} in the legume *P. officinalis*, the mangrove *A. germinans*, and the herbaceous *C. erubescens* and *M. arborescens*. Phosphorus showed a similar species distribution pattern, being below 20 mmol kg^{-1} in both *Clusia* species, *L. guianensis*, and in *C. icaco*. The distribution of concentrations of S showed that most species had concentrations between 60 and 110 mmol kg^{-1}, the lowest concentrations, below 50 mmol kg^{-1}, were recorded for *M. flexuosa* and *B. serrulatum*, and the highest concentrations, above 150 mmol kg^{-1}, corresponded to another fern *A. aureum*, *M. arborescens*, and the sample of

Table 3 Concentrations of the elements N, S, and P in the organic matter of leaves from species sampled along the Guanoco transect

Distance m	Species	N (mmol kg^{-1})	S (mmol kg^{-1})	P (mmol kg^{-1})	N/P	N/S
0	*Avicennia germinans*	2,164	113	71	31	19
	Rhizophora racemosa	1,193	63	26	47	19
50	*Crinum erubescens*	1,593	114	83	19	14
	Pterocarpus officinalis	2,200	69	32	68	32
	Rhizophora racemosa	993	70	27	37	14
300	*Pterocarpus officinalis*	2,621	80	41	64	33
	Rhizophora racemosa	879	99	25	35	9
500	*Pterocarpus officinalis*	1,807	61	32	56	30
	Rhizophora racemosa	993	69	29	34	14
650	*Acrostichum aureum*	957	150	23	41	6
	Acrostichum danaefolium	1,050	89	36	29	12
	Laguncularia racemosa	1,200	91	48	25	13
	Montrichardia arborescens	2,943	172	75	39	17
	Rhizophora racemosa	900	83	27	34	11
750	*Lagenocarpus guianensis*	450	84	10	47	5
850	*Blechnum serrulatum*	1,007	58	20	50	17
	Chrysobalanus icaco	707	83	11	62	8
	Clusia sp. 1	579	126	12	46	5
	Clusia sp. 2	579	101	11	51	6
	Fuirena umbellata	1,000	96	18	54	10
	Tabebuia insignis	700	211	17	41	3
1,250	*Blechnum serrulatum*	979	61	27	37	16
	Chrysobalanus icaco	800	94	15	52	9
	Clusia sp. 2	493	57	14	36	9
	Tabebuia insignis	879	69	20	43	13
1,500	*Blechnum serrulatum*	829	44	16	51	19
	Chrysobalanus icaco	857	76	15	55	11
	Clusia sp. 1	614	70	14	44	9
	Lagenocarpus guianensis	564	60	9	63	9
	Mauritia flexuosa	671	32	15	44	21
	Tabebuia insignis	914	82	20	46	11
	Average	1,100	88	27	45	14

T. insignis from site 6. In general the N and P concentrations were highly correlated (N = 395.9 + 25.98 P; $r^2_{adj} = 0.60$, $P < 0.0001$), dispersion of values increasing at N concentrations above 1,500 mmol kg^{-1}. The species *C. erubescens*, *A. germinans*, *M. arborescens* always had comparatively high levels of N, P, and S.

The regression analysis of the N, P, and S concentrations of all species against distance from river fringe showed significant reductions of N and P (r^2_{adj} for N = 0.289, $P = 0.001$; r^2_{adj} for P = 0.323, $P = 0.0005$), whereas S remained constant.

Stable isotopes ratios in leaves and soils

The species sampled had carbon isotope ratios ranging from −25.5 to −32‰ (average −28.3‰). The highest values corresponded to the ferns species, *M. arborescens* and the samples of *R. racemosa* collected from sun-exposed branches. The more-negative values

Table 4 Nitrogen and carbon isotopic ratios in plant leaves and soil samples along the Guanoco transect

Distance (m)	Species	Plant leaves		Soils	
		δ ^{15}N (‰)	δ ^{13}C (‰)	δ ^{15}N (‰)	δ ^{13}C (‰)
0	*Rhizophora racemosa*	−0.38	−28.23		
0	*Avicennia germinans*	1.39	−30.16		
50	*Rhizophora racemosa*	3.35	−29.92	2.16	−28.14
50	*Pterocarpus officinalis*	−1.16	−28.93		
50	*Crinum erubescens*	1.59	−32.06		
300	*Rhizophora racemosa*	2.27	−30.64	1.32	−27.46
300	*Pterocarpus officinalis*	−1.15	−30.48		
500	*Rhizophora racemosa*	3.96	−25.42	0.43	−28.64
500	*Pterocarpus officinalis*	−1.33	−29.23		
650	*Rhizophora racemosa*	2.16	−25.87	0.28	−28.74
650	*Acrostichum danaefolium*	3.51	−25.62		
650	*Montrichardia arborescens*	3.58	−25.40		
650	*Laguncularia racemosa*	1.42	−28.27		
650	*Acrostichum aureum*	2.15	−26.11		
750	*Lagenocarpus guianensis*	2.49	−27.78	1.14	−28.43
850	*Clusia* sp. 1	−7.77	−27.58	−1.95	−27.94
850	*Fuirena umbellata*	−0.05	−28.38		
850	*Tabebuia insignis* var. *monophylla*	3.45	−27.89		
850	*Chrysobalanus icaco*	−1.78	−30.52		
850	*Clusia* sp. 2	−7.50	−27.40		
850	*Blechnum serrulatum*	1.19	−26.24		
1,250	*Clusia* sp. 2	−9.09	−27.90	−3.27	−28.28
1,250	*Tabebuia insignis* var. *monophylla*	−1.37	−29.88		
1,250	*Blechnum serrulatum*	−2.68	−29.13		
1,250	*Chrysobalanus icaco*	−6.89	−30.25		
1,500	*Mauritia flexuosa*	−2.03	−27.59	−0.73	−28.2
1,500	*Clusia* sp. 1	−10.55	−28.49		
1,500	*Chrysobalanus icaco*	−7.25	−30.78		
1,500	*Lagenocarpus guianensis*	0.32	−27.76		
1,500	*Tabebuia insignis* var. *monophylla*	−8.97	−27.30		
1,500	*Blechnum serrulatum*	−4.00	−26.62		
	Averages	−1.33	−28.32	−0.08	−28.23

corresponded to *C. erubescens*, a plant from the forest floor, *P. officinalis*, and *C. icaco*. The distribution of carbon isotope ratios along the transect did not show any particular trend (Table 4). Soil samples were also very similar throughout the transect, averaging −28.2‰.

The range of nitrogen isotope ratios was larger than that of the carbon isotope ratios, +4 to −10.5‰, averaging −1.4‰. All the mangrove species (*A. germinans*, *L. racemosa*, and *R. racemosa*), the ferns *A. aureum* and *A. danaefolium*, and the aroid *M. arborescens* had values above average and positive. The samples of *P. officinalis* were always negative and similar to the average, while those of *C. icaco*, and the *Clusia* species were always quite negative. The values of *T. insignis* decreased from 3.5‰ at site 7, to 1.4‰ at site 8, and −9‰ at site 9. The distribution of nitrogen isotope ratios of leaves and soils along the transect showed a tendency for more positive values obtained from samples towards

Table 5 Analysis of variance of the composition of plant samples separated into those belonging to the mangrove belt (I, 0–700 m from the river fringe) and the back swamp (II, 700–1,500 m from the river fringe). Element concentrations in mmol kg^{-1}. As the Bartlett test showed unequal variances for several parameters, a Welch's ANOVA allowing for unequal standard deviations was performed (JMP Statistical Software Package, SAS Institute 2002)

Component	I	II	F	Prob >*F*	Bartlett Test	$F_{u.v.}$	Prob > $F_{u.v.}$
Plant samples							
S	95	83	0.79	0.382	0.492	0.82	0.373
N*	1,535	742	20.52	<0.001	<0.001	17.21	<0.001
P*	41	16	25.16	<0.001	<0.001	20.97	<0.001
K	328	200	4.28	0.048	<0.001	3.61	0.077
Na*	213	88	6.57	0.016	0.001	5.68	0.029
Mg	129	93	4.22	0.049	0.647	4.11	0.053
Ca	262	220	0.60	0.445	0.562	0.62	0.438
% Ash*	8.2	6.3	5.46	0.027	0.199	5.11	0.034
∑Cation*	932	601	13.84	<0.001	0.048	12.55	0.002
δ ^{15}N ‰*	1.5	−3.7	16.80	<0.001	0.003	19.32	<0.001
δ ^{13}C ‰	−28.3	−28.3	<0.01	0.983	0.054	<0.01	0.984
Soil samples							
δ ^{15}N ‰*	1.1	−2.3	26.15	0.001	0.690	23.33	0.005
δ ^{13}C ‰	−28.3	−28.2	0.211	0.660	2.857	0.25	0.633

*Indicates significant differences at $P \leq 5\%$

the river coast line, and more negative values found towards the forest and herbaceous swamps (Table 4).

Discussion

Total cation concentration in substrate and interstitial water showed a clear reduction from the river fringe to the back swamps, whereas the organic matter increased markedly in the same direction. The soil in the back swamps is organic, consisting of true peat soils. This pattern is probably associated with the flooding regime. The mangrove belt on the natural levee (sites 1 to 4) is flooded with variable intensity, once or twice per day, while the back swamps have a more-permanent flooding regime, leading to the accumulation of organic matter that cannot decompose due to hypoxia. The cation concentration of the interstitial water decreased quickly after site 4, indicating its association with mineral components. The increase in Na concentration at site 5 is probably related to the end of the direct tidal influence, and may explain the presence there of the *Laguncularia* belt. Site 5 is located at the back of the mangrove belt, where the soil changes from a silt clay sulfaquent with up to 18 mol C kg^{-1}, to a typic tropofibist, and the influence of daily tides disappears (MARNR 1990). At this site, the opening of the canopy generates higher evapotranspiration rates, resulting in higher Na concentration. The vegetation belt parallel to the river between the river fringe and 650 m therefore constitutes a mangrove belt, where salinity allows the establishment and reproduction of true halophytes. Within this belt *A. germinans* is restricted to the first few meters from the fringe, whereas *L. racemosa* is restricted to the back of the belt, possibly because this species is not able to compete for light with the taller, more-competitive *R. racemosa*. Beyond the 650 m line, geomorphologic depressions lead to the formation of back swamps, flooded mainly by rainfall, their drainage being prevented by their location behind the natural levee and the dam effect of tides. Echezuría et al. (2002) and White et al. (2002) described environmental settings of this type that are frequent in the Orinoco Delta.

Cation concentration in leaf tissues revealed species-specific patterns, which included cation accumulators and excluders (Table 2). Within these species groups the Na-excluding character of *P. officinalis*, and the large K accumulation capacity of *C. erubescens* are noteworthy. These properties

allow these species to coexist successfully with mangroves in saline soils under high rainfalls. Mangrove species differ in their cation accumulation pattern. *Rhizophora racemosa* is a relative Ca accumulator, whereas *A. germinans* is characterized by very low levels of Ca, certainly related to the production of oxalate in root and leaf tissues (Medina and Francisco 1997; Medina et al. 2001). *Avicennia germinans* behaves as a relative Mg accumulator, confirming previous reports from other mangrove communities under contrasting rainfall regimes (Medina et al. 2001; Barboza et al. 2006). The physiological significance of Ca versus Mg accumulation in mangroves remains a subject for experimental study.

The δ ^{13}C values were within the range of C3 plants. Differences among samples appeared to be related more to light conditions and the position within the canopy than to water relations. The more-negative values corresponded to leaves obtained from partially shaded branches, growing in the lower canopy or near the forest floor, as in the case of *C. erubescens*. This would be expected from the Farquhar et al. (1982) model relating carbon isotope discrimination to the internal CO_2 concentration. Lower light intensity reduces photosynthetic CO_2 uptake, favoring discrimination by RuBP-carboxylase. Besides, in the lower canopy CO_2 derived from soil respiration is depleted in ^{13}C and its fixation by lower canopy leaves results in more-negative ^{13}C ratios (Medina and Minchin 1980).

The N isotope ratios showed an unusually wide range for a natural setting and a definite decreasing trend along the transect. Fry et al. (2000) reported large variations in δ ^{15}N values in *Rhizophora mangle* in Florida, and attributed the frequency of high positive values to the influence of anthropogenic sources of nitrogen. As there was no anthropogenic pollution in our study site, the cause of this variation was probably associated with properties of the nitrogen cycle and lower N demands of plants as a result of P limitation. During the decomposition of organic matter the isotopic signature of N remains constant. Subsequently processes of denitrification, NH_3 volatilization, and leaching of mineral N lead to ^{15}N enrichment in the soil. Plants using N from these ^{15}N-enriched upper soil layers also become enriched in this isotope. Therefore, it was expected that soils and vegetation influenced by bidiurnal tides, with active transport of sediments, and transient periods of oxygenation favoring denitrification, had to be relatively enriched in ^{15}N, whereas areas separated from sediment sources, where organic matter decomposition is restricted by hypoxia, had to be characterized by relatively negative N isotope ratios. This distinction was expected to be reinforced by the limitation of P supply in the backswamps. Phosphorus limitation reduces N demand for plant growth, thereby increasing the discrimination against the heavy isotope during uptake of mineral N. In a tree height gradient within a mangrove community in Belize McKee et al. (2002) conducted differential fertilization experiments and showed that P availability regulates the utilization of available N. These authors reported that tree height and leaf δ ^{15}N values were reduced by lower P availability, as indicated by lower leaf P concentration. Fertilization with P of the dwarf mangrove community resulted in the production of leaves with similar δ ^{15}N values as the mangrove community not limited by P.

A synthesis of the findings of this paper can be attained by separating the leaf and soil samples into the river mangrove belt and the back swamp. A one-way analysis of variance (ANOVA) of these groups shows clear-cut differences in nutritional conditions (Table 5). Among the cations, only Na showed a significant reduction in leaf tissues of back swamp species. This was expected from the measured Na concentration in interstitial water. The concentrations of K and Mg were on average lower in the back-swamp species, but the ANOVA showed only marginal significance.

Carbon and sulfur concentrations, and δ ^{13}C values were nearly identical for the two groups of plants, whereas N and P, and δ ^{15}N values showed highly significant differences. The conclusion from this analysis is that back swamp plants may be limited by the availability of N, determined by the slow rate of organic matter decomposition, and P due to the isolation of the swamp from the source of sediments. Similar nutritional constraints have been described for mangrove communities in Australia (Boto and Wellington 1983), Belize (Feller 1995; Feller et al. 2003), and Florida (Koch and Snedaker 1997). The δ ^{15}N values of the mangrove belt along the Guanoco River were more positive than those reported by McKee et al. (2002), but lower than those reported by

Medina et al. (2001) from mangrove communities in the Bragança Peninsula in Brazil.

Our results support the nutrient availability hypothesis, indicating sufficient nutrient supply along the river fringe, and nutrient limitation, particularly N and P, in the back swamps. They also suggest that vegetation zonation is the result of several superimposed gradients of hydroperiod, salinity, and nutrient availability. Results also indicate that relative accumulation of ^{15}N in these systems derives from the relative openness of the N cycle, as discussed by Martinelli et al. (1999).

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