Nutritional Efficiency of Forest Species in Natural Regeneration of Tropical Forest in Brazil

Roseane K. S. da Silva[1], Ana L. P. Feliciano[1], Luiz C. Marangon[1], Maria Betânia G. dos S. Freire[2], Fernando J. Freire[2], Rosival B. de A. Lima[1] & Ane C. F. da Silva[3]

[1]Forest Science Department, Federal Rural University of Pernambuco, Recife, Brazil

[2]Agronomy Department, Federal Rural University of Pernambuco, Recife, Brazil

[3]Princess Izabel Campus, Federal Institute of Paraíba, Princesa Izabel, Brazil

Correspondence: Fernando J. Freire, Agronomy Department, Federal Rural University of Pernambuco, Recife, Rua Dom Manoel de Medeiros, CEP 52171-900, Brazil. Tel: 55-81-3320-6223. E-mail: fernando.freire@ufrpe.br

Abstract

The knowledge of the nutritional aspects of native species, mainly in natural regeneration, may be important for understanding their establishment, particularly in areas with low nutrient availability soils, such as tropical soils. This study aimed to determine the biological utilization efficiency (BUE) of the nutrients N, P, K, Ca, and Mg of forest species of natural regeneration in a Lowlands Dense Ombrophilous forest fragment in Pernambuco, Brazil. A phytosociological study of the fragment was carried out and were defined the ten species with the highest absolute density (AD). Three individuals per species were selected. The N, P, K, Ca, and Mg contents were determined in the sample leaves of the species, and the foliar biomass was determined "*in loco*". Nine individuals of each species were collected according to the following diameter intervals at the base (DBs): DBs<5 cm; 5≤DBs<10 cm and 10≤DBs<15 cm. The content, stock and BUE of nutrients were calculated per species. The BUE of nutrients by species varied according to the following decreasing order: P>Mg>K>Ca>N. The highest BUE of nutrients was of the species *Protium heptaphyllum*. In tropical soils of low natural fertility, the use of these species can be recommended in environmental reforestation projects. The difference in the nutritional demand of the forest species can indicate the planting of those with greater capacity of absorption and BUE of nutrients, being more efficient in areas of soils with low natural fertility like in the tropical forests.

Keywords: forest nutrition, nutrient content, nutrient stock, biological utilization efficiency

1. Introduction

The Atlantic Forest is responsible for a significant portion of Brazil's biological diversity and has been impacted by constant anthropogenic pressures, endangering the richness of endemic species that make up the biome (Bosa, Pacheco, Pasetto, & R. Santos, 2015). According to A. Chaves, R. M. S. Santos, J. O. Santos, Fernandes, and Maracajá (2013), due to the high degree of anthropogenic disturbances in this biome, its conservation represents one of the greatest challenges of the Brazilian tropical regions.

Natural regeneration represents the interaction of natural processes of reestablishment of the forest ecosystem. It is part of the forest growth cycle and refers to the initial stages of its establishment and development (Gama, Botelho, & Bentes-Gama, 2002). It is considered the intermediate stage between the seedling and the adult vegetative or reproductive stage, fundamental for the maintenance of forest balance, since the failure of adaptive processes during this period may make it difficult to establish or even eliminate the species from the site (Amo-Rodríguez & Gómez-Pompa, 1976).

The biological utilization efficiency (BUE) of nutrients expresses the ability of plants to absorb and utilize nutrients (Barros & Novais, 1990), being considered an efficient plant those that produce the maximum biomass per absorbed nutrient (Stahl, Ernani, Gatiboni, D. Chaves, & Neves, 2013). Many factors contribute to the BUE of nutrients of the plants, such as: associations with beneficial fungi or bacteria (Jacoby, Peukert, Succurro, Koprivova, & Kopriva, 2017); nature of soils (Baligar, Fageria, & He, 2007); and taxonomic group e.g. Leguminosae (Fabaceae) (Baribault, Kobe, & Finley, 2012).

The evaluation of the BUE of nutrients of different forest species is an important parameter to assist in the choice

of material to be used in reforestation (Caldeira, Rondon Neto, & Schumacher, 2002); however, the studies that have been carried out emphasize mainly commercial species (Stahl *et al*., 2013; Batista, Furtini Neto, & Deccetti, 2015).

Information on BUE of nutrients of native forest species is scarce in literature, and when they exist, they refer only to the adult arboreal stratum, as in the works of Espig *et al*. (2008) and B ündchen, Boeger, Reissmann, and S. L. C. Silva (2013), which evaluated the BUE of nutrients in adult tree species in forest formations of the Atlantic Forest Biome. No studies were found to evaluate species in the natural regeneration phase. Young individuals (seedlings) are usually evaluated in experiments in a protected and controlled environment, with the objective of studying the BUE of nutrients in response to the fertilization of a certain nutrient, such as the studies developed by N. Souza *et al*. (2012) and N. Carnevali, Marchetti, Vieira, T. Carnevali, and Ramos (2016).

The distribution of mineral nutrients in the plant and its components is not homogeneous. However, the crow's biomass, although it represents a small part of the total biomass of the tree, has a high mineral nutrient content. It is concentrated mainly in the leaves, and is even higher in the initial stages of plant growth. Thus, the determination of nutrient content in the leaves is the most used way to evaluate their stock (A. C. Silva, A. R. Santos, & Paiva, 1998), if the biomass is measured or estimated. Therefore, with the stock and the biomass one can calculate the BUE of nutrients of the species of a forest stand (Espig *et al*., 2008).

The knowledge of nutritional aspects of native species is important in order to understand the establishment of these species in their origin sites, especially in areas with low nutrient availability soils, such as tropical soils (N. Souza *et al*., 2012). This information, when obtained specifically for natural regeneration species, which are in the initial phase of its development (Felfili, Rezende, Silva-J únior, & M. A. Silva, 2010), combined with the knowledge of these species' autecology (Schorn & Galv ão, 2006) may subsidize the indication for use in reforestation.

The hypothesis of this study is that in nutritionally balanced forests, BUE of nutrientes should vary between species by nutrient. The most efficient species to use N is not the same as most efficient to use P.

In this context, this work aimed to determine the BUE of the nutrients N, P, K, Ca, and Mg of forest species with the highest absolute density (AD) of natural regeneration in a Lowlands Dense Ombrophilous forest fragment.

2. Method

The study was carried out in a Lowlands Dense Ombrophilous forest fragment (L. Martins & Cavararo, 2012), of approximately 79 ha, in the municipality of Sirinha ém, Pernambuco, Brazil, under the coordinates UTM 25L 259089 and 9053293; 259604 and 9053741; 259727 and 9052723; 259920 and 9052956, with an mean altitude of 63 m. The region presents an Am monsoon climate, according to Köppen's classification with a mean annual temperature of 25.6 ℃ (Alvares, Stape, Sentelhas, Gon çalves, & Sparovek, 2013).

The rainfall data of the Pernambuco State Agency for Water and Climate - APAC recorded an mean annual rainfall of around 1,800 mm (P. Oliveira *et al*., 2016). Soils found in the region are of Yellow Latosol, Yellow Argisol, Red-Yellow Argisol, Gray Argisol, Gleissol, Cambisol, and Fluvic Neosols (H. G. Santos *et al*., 2013).

For soil chemical characterization, particle size distribution and soil textural class of the forest fragment, four simple samples were collected and homogenized, giving rise to a composite sample. They were sampled in 40 plots (10 m x 25 m) that were distributed systematically in the fragment. Samples were collected at two depths (0.0-0.10 m and 0.10-0.20 m) (Table 1).

Table 1. Soil chemical attributes particle size distribution and soil textural class in the Lowlands Dense Ombrophilous forest fragment, Pernambuco, Brazil

Soil attribute	Depth (m)	
	0.0-0.10	0.10-0.20
pH (H_2O)	3.88 ± 0.23	4.15 ± 0.23
P (mg dm^3)	1.33 ± 0.52	1.20 ± 0.40
Ca^{2+} ($cmol_c$ dm^{-3})	0.47 ± 0.21	0.22 ± 0.13
Mg^{2+} ($cmol_c$ dm^{-3})	0.64 ± 0.32	0.52 ± 0.23
K^+ ($cmol_c$ dm^{-3})	0.07 ± 0.04	0.05 ± 0.03
Al^{3+} ($cmol_c$ dm^{-3})	1.41 ± 0.36	1.22 ± 0.25
(H+Al) ($cmol_c$ dm^{-3})[1]	6.10 ± 1.75	4.68 ± 1.39
TOC (g kg^{-1})[2]	25.2 ± 0.88	18.0 ± 0.53
SB[3]	1.18 ± 0.39	0.79 ± 0.29
CEC$_{effective}$ ($cmol_c$ dm^{-3})[4]	2.59 ± 0.42	2.01 ± 0.32
CEC$_{potential}$ ($cmol_c$ dm^{-3})[5]	7.28 ± 1.74	5.47 ± 1.36
m (%)[6]	54.44 ± 11.85	60.70 ± 10.74
V (%)[7]	16.21 ± 7.09	14.44 ± 7.25
Total Sand (g kg^{-1})	481.60 ± 6.96	432.90 ± 5.50
Coarse Sand (g kg^{-1})	384.80 ± 6.46	335.90 ± 4.91
Fine Sand (g kg^{-1})	96.80 ± 1.31	97.10 ± 1.58
Silt (g kg^{-1})	252.70 ± 6.21	270.80 ± 8.54
Clay (g kg^{-1})	265.70 ± 4.95	296.30 ± 7.70
Textural class	Sandy clay loam	Loam clay

[1]Potential acidity; [2]Total organic carbon; [3]Sum of bases; [4]Effective cation exchange capacity; [5]Potential cation exchange capacity; [6]Saturation by aluminum; [7]Base saturation.

The Ca^{2+}, Mg^{2+} and Al^{3+} were extracted by 1.0 mol L^{-1} KCl solution and determined by titration. P, K^+, Fe, Cu, Zn and Mn were extracted by Mehlich-1 solution. P was determined by spectrophotometry, K^+ by flame photometry and Fe, Cu, Zn and Mn by atomic absorption spectrophotometry. Potential acidity (H+Al) was extracted by 0.5 mol L^{-1} calcium acetate solution and determined by titration, and the total organic C (TOC) determination was performed by oxidation using the K dichromate method. With the results of these chemical analyzes, the sum of bases (SB), base saturation (V), saturation by Al (m), effective cation exchange capacity (CEC$_{effective}$), and potential cation exchange capacity (CEC$_{potential}$) were all calculated (Donagema, Campos, Calderano, W. Teixeira, & Viena, 2011).

For the sampling of shrub-tree species of natural regeneration, 40 subunits of 25 m² (5 x 5 m) were systematically allocated. These subunits were implemented on the right side of 40 sampling units of 250 m² (10 x 25 m) (Figure 1), previously permanently allocated for the study of the floristic composition of the adult shrub-tree community, equidistant by 25 m and interspersed to the right and left. The fragment is a permanent preservation area in accordance with the Brazilian legislation of an agricultural enterprise producing sugarcane.
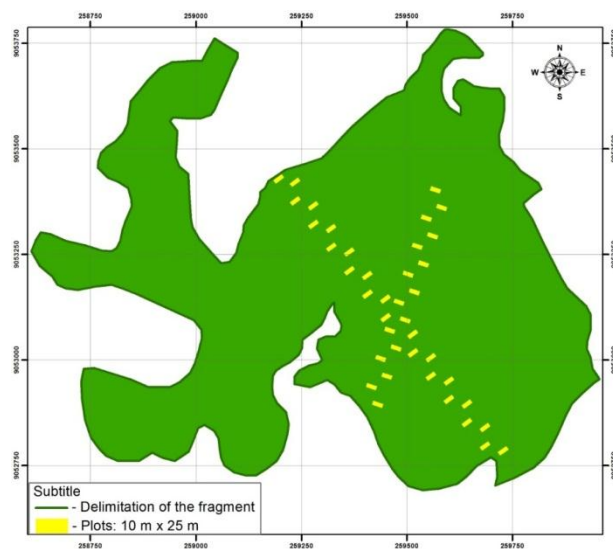

Figure 1. Schematic diagram of the plots distribution in the Lowlands Dense Ombrophilous forest fragment, Pernambuco, Brazil

Natural regeneration studies were established based on the inclusion level, proposed by Finol (1971) adapted by Marangon (1999), in which individuals who presented a diameter at breast height (DBH) < 15 cm and height ≥ 1 m were measured. Due to the height inclusion level, the diameter measurements were performed at the base, 30 cm from the ground (DBs).

Sampling of the leaves was performed between 6 and 9 h am during the hottest and humid period of the year. Sampling of the species was also performed in individuals with good phytosanitary status in shrub-tree with better morphological performance in the forest fragment.

The species identification was according to the Angiosperm Phylogeny Group - APG III (2009) classification system. With the data, the ten species of natural regeneration with the highest AD were defined using the following expression (M üeller-Dombois & Ellemberk, 1974):

$$AD = Ni/A$$

Where: AD is the absolute density (ind. ha^{-1}); N is the number of individuals of i species; and A is the sample area in hectares. The sampled individuals were counted in 40 plots of 5 x 5 m totaling 1,000 m^2. Subsequently the data were estimated to 10,000 m^2 (one hectare).

The ten forest species with the highest AD in natural regeneration of the fragment were classified as Ecological Group (EG) (Table 2). The EG classification followed the proposal of Gandolfi, Leit ão Filho, and Bezerra (1995), who defined as: Pioneers (P) – light dependent species; Initial secondary (IS) – species that occur in conditions of medium shading or not very intense luminosity; and Late secondary (LS) – species that develop in the understory under light or dense shade conditions, and can remain in this environment for a lifetime or grow until reaching the canopy or emergent condition.

Table 2. Species with the highest absolute density (DA) in natural regeneration in the Lowlands Dense Ombrophilous forest fragment

Species	Family	Ecological group	Absolute density (Ind. ha^{-1})	DBs[3] (cm)
Brosimum rubescens Taub.	Moraceae	IS[1]	1,500 ± 82	3.2 ± 1.4
Thyrsodium spruceanum Benth.	Anacardiaceae	IS	580 ± 36	4.6 ± 2.8
Tovomita mangle G. Mariz	Clusiaceae	IS	560 ± 21	4.9 ± 2.5
Anaxagorea dolichocarpa Sprague & Sandwith	Annonaceae	LS[2]	340 ± 14	6.0 ± 3.7
Eschweilera ovata (Cambess.) Miers	Lecythidaceae	IS	340 ± 13	7.9 ± 2.0
Protium arachouchini March.	Burseraceae	LS	280 ± 11	6.4 ± 3.2
Caraipa densifolia Mart.	Calophyllaceae	IS	280 ± 18	4.7 ± 2.5
Talisia retusa R.S. Cowan	Sapindaceae	IS	260 ± 11	5.2 ± 3.2
Inga capitata Desv.	Fabaceae	IS	250 ± 10	5.2 ± 2.0
Protium heptaphyllum (Aubl.) Marchand	Burseraceae	IS	240 ± 10	6.7 ± 3.7

[1]Initial secondary; [2]Late secondary; [3]Diameter at base (medium).

For the foliar sampling, there were selected three individuals of each species with highest AD that presented DBs similar to the mean DBs of all individuals of the species. They were also selected for their phytosanitary status and good cup formation, which properly characterizes the species.

Fifteen leaves in the middle third of the plant were collected. The collected leaves were packed in plastic bags and stored in styrofoam boxes with ice. Subsequently, the plastic bags were replaced by paper bags and then taken to a forced air circulation chamber at 65 ℃ in order to dry the foliar material. After reaching constant weight, the material was milled, homogenized, and conditioned in previously cleaned and dried vials, for further analysis.

The nutrients P, K, Ca, and Mg were extracted by nitric-perchloric digestion (Bataglia, Furlani, J. Teixeira, Furlam, & Gallo, 1983). Next, Ca and Mg were determined by atomic absorption spectrophotometry, P dosed by colorimetry (Braga & Defelipo, 1974), and K determined flame photometry. N was extracted by sulfur-digestion and determined by distillation and titration by the Kjeldahl method (Tedesco, Gianello, Bissani, & Bohnen, 1995).

The mean fresh foliar biomass of each species (g plant^{-1}) was determined by collecting all leaves of three individuals, according to the DBs intervals: DBs<5 cm; 5≤DBs<10 cm and 10≤DBs<15 cm, totaling nine individuals per species. The leaves of each individual were weighed in the field to obtain the fresh matter weight.

The percentage of moisture of each species was determined by weighing three subsamples of variable size

($\geq$100 g) and subjected to a temperature of 65 ℃ to constant weight. Dry foliar biomass per species was determined by the fresh and dry weight ratio of the samples. To calculate the total dry biomass of each species per unit area (kg ha^{-1}), the mean dry biomass of the species was multiplied by AD (ind. ha^{-1}).

The nutrient stock in the foliar biomass of the species in kg ha^{-1} was calculated by multiplying the nutrient content (g kg^{-1}) by the foliar biomass (kg ha^{-1}). The BUE of nutrients was calculated as the ratio of the foliar biomass of the species and the nutrient stock in the biomass (Espig *et al*., 2008).

The statistical procedure used to study the data of content, stock, and BUE of the nutrients N, P, K, Ca, and Mg was the analysis of variance (ANOVA). Comparisons of means among the species using Scott-Knott's test at 5% probability were also used, when the effects were significant by the F test at 5% probability level. The Kolmogorov-Smirnov test was used to test the hypothesis of data normality (Fisher, 1990). The SAS software was used for the statistical analyses (Statistical Analysis System, version 8.2, SAS Institute, Cary, N.C., USA).

3. Results

The total foliar biomass by shrub-tree species of natural regeneration in the Lowlands Dense Ombrophilous forest fragment varied more than seventeen times between the species of lower biomass per area (*C. densifolia*) and those of highest biomass (*B. rubescens*) (Table 3).

Table 3. Foliar biomass per individual (g plant $^{-1}$) and per area (kg ha^{-1}) of the forest species with highest absolute density (DA) in natural regeneration in the Lowlands Dense Ombrophilous forest fragment

Forest species	Foliar biomass per individual (g plant^{-1})	Foliar biomass per area (kg ha^{-1})
Brosimum rubescens	181.54 ±87.39	272.31 ±131.08
Thyrsodium spruceanum	66.30 ±35.53	38.46 ±20.61
Tovomita mangle	127.27 ±43.08	71.27 ±24.12
Anaxagorea dolichocarpa	94.62 ±20.14	32.17 ±6.85
Eschweilera ovata	69.17 ±28.18	23.52 ±9.58
Protium arachouchini	289.29 ±74.40	81.00 ±20.83
Caraipa densifolia	55.88 ±19.88	15.65 ±5.57
Talisia retusa	145.06 ±14.31	37.72 ±3.72
Inga capitata	63.42 ±16.54	15.86 ±4.13
Protium heptaphyllum	403.60 ±212.24	96.86 ±50.94
Total foliar biomass	-	684.81 ±27.74

P. heptaphyllum and *P. arachouchini* were the species that presented the largest individual biomasses, possibly because they present composite leaves with larger dimensions and *C. densifolia* was the species that presented the lowest foliar biomass.

Foliar biomass per individual ranged from 55.88 to 403.60 g plant^{-1} (Table 3). Despite the great difference between individual biomasses, it was verified that the total biomass per species is directly related to AD, because *B. rubescens*, even though it did not present the highest individual biomass, was highlighted due to the high AD (Table 2).

The nutrient content in the leaves of natural regeneration species, on mean, was distributed according to the following decreasing order: N>Ca>K>Mg>P (Table 4).

Table 4. Nutrient content (g kg^{-1}) and stock (kg ha^{-1}) in the species with highest absolute density (DA) in natural regeneration in the Lowlands Dense Ombrophilous forest fragment

Forest species	N	P	K	Ca	Mg
			g kg^{-1}		
Brosimum rubescens	15.31 ±0.70b	1.47 ±0.16b	3.27 ±0.77b	5.05 ±0.82b	2.98 ±0.23b
Thyrsodium spruceanum	15.63 ±1.68b	1.38 ±0.15b	2.41 ±0.24b	6.76 ±2.12b	3.94 ±0.28a
Tovomita mangle	17.03 ±1.53b	1.33 ±0.13b	5.27 ±0.54b	12.20 ±2.22a	4.18 ±0.60a
Anaxagorea dolichocarpa	17.55 ±0.82b	2.45 ±0.11a	8.14 ±1.76a	14.31 ±0.96a	3.98 ±0.24a
Eschweilera ovata	18.71 ±1.90b	1.34 ±0.09b	4.33 ±1.12b	4.73 ±0.76b	3.42 ±0.62b
Protium arachouchini	15.59 ±1.54b	1.37 ±0.11b	4.84 ±0.98b	5.01 ±0.86b	1.67 ±0.24c
Caraipa densifolia	15.59 ±0.53b	1.51 ±0.18b	3.66 ±0.41b	3.58 ±0.49b	1.61 ±0.09c
Talisia retusa	15.03 ±0.9b	1.47 ±0.32b	5.16 ±2.45b	10.76 ±3.81a	4.29 ±0.55a
Inga capitata	23.85 ±2.71a	1.35 ±0.12b	4.29 ±0.17b	4.42 ±3.71b	2.19 ±0.13c
Protium heptaphyllum	15.17 ±2.04b	1.27 ±0.07b	3.11 ±1.07b	3.24 ±1.18b	0.98 ±0.30d
Mean	16.94	1.49	4.45	7.01	2.92
F$_{calculated}$	80.80**	13.80**	5.60**	11.00**	31.22**
CV (%)[1]	9.34	10.74	26.27	29.42	12.96
			kg ha^{-1}		
Brosimum rubescens	4.17 ±0.19a	0.40 ±0.04a	0.89 ±0.21a	1.37 ±0.22a	0.81 ±0.06a
Thyrsodium spruceanum	0.60 ±0.06d	0.05 ±0.005d	0.09 ±0.005c	0.26 ±0.08c	0.15 ±0.01c
Tovomita mangle	1.22 ±0.10c	0.10 ±0.01c	0.37 ±0.04b	0.87 ±0.15b	0.30 ±0.04b
Anaxagorea dolichocarpa	0.56 ±0.02d	0.08 ±0.005c	0.26 ±0.05b	0.46 ±0.03c	0.13 ±0.01c
Eschweilera ovata	0.44 ±0.04e	0.03 ±0,00e	0.10 ±0.02c	0.11 ±0.02d	0.08 ±0.01d
Protium arachouchini	1.26 ±0.12c	0.11 ±0.01b	0.39 ±0.08b	0.41 ±0.06c	0.14 ±0.02c
Caraipa densifolia	0.24 ±0.01e	0.02 ±0.005e	0.06 ±0.005c	0.05 ±0.005d	0.03 ±0.005e
Talisia retusa	0.57 ±0.03d	0.06 ±0.01d	0.20 ±0.08c	0.41 ±0.14c	0.16 ±0.02c
Inga capitata	0.38 ±0.04e	0.02 ±0,00e	0.06 ±0.005c	0.07 ±0.05d	0.03 ±0.005e
Protium heptaphyllum	1.47 ±0.19b	0.12 ±0.005b	0.30 ±0.10c	0.31 ±0.11c	0.09 ±0.03d
Mean	1.09	0.10	0.27	0.43	0.19
F$_{calculated}$	353.86**	144.13**	24.82**	39.79**	198.14**
CV (%)	9.78	16.29	31.84	25.87	14.82

[1]Coefficient of variation = Standard deviation/Mean x 100.

Means followed by equal letters in the columns do not differ from each other at 5% probability level by the Scott-Knott's test.

** Significant at 1% probability by the F test. [ns]Not significant.

The N content in the leaves ranged from 15.03 g kg^{-1} to 23.85 g kg^{-1}, with a mean of 16.94 g kg^{-1} (Table 4). The species with the highest N content was *I. capitata*, the only leguminous species of the group of ten of greater DA (Table 2).

The Ca content was higher in the leaves of *A. dolichocarpa*, *T. mangle* and *T. retusa*. A second group with lower Ca content was formed by *T. spruceanum, B. rubescens, P. arachouchini, I. capitata, C. densifolia*, and *P. heptaphyllum* (Table 4). The Ca content in these species ranged from 3.24 to 14.31 g kg^{-1}, with a mean of 7.01 g kg^{-1}.

K content in the leaves of the studied species ranged from 2.41 g kg^{-1} to 8.14 g kg^{-1} and presented a mean of 4.45 g kg^{-1}, especially *A. dolichocarpa*, which presented the highest content of this nutrient (Table 4).

P. heptaphyllum showed the lowest content of Mg, which was 0.98 g kg^{-1}, while in the species *T. retusa*, *T. spruceanum*, *T. mangle* and *A. dolichocarpa* the Mg content was higher (Table 4).

P content foliar biomass of the species varied from 1.33 to 2.45 g kg^{-1}, with a mean of 1.49 g kg^{1}. P content in *A. dolichocarpa* was higher than of the other species (Table 4).

Nutrient stock in the foliar biomass of natural regeneration species followed the decreasing order: N>Ca>K>Mg>P. This decreasing sequence varied according to species: *T. spruceanum* was N>Ca>Mg>K>P; *C. densifolia* was N>K>Ca>Mg>P; and *P. heptaphyllum* was N>Ca>K>P>Mg (Table 3). The highest nutrient stock were observed in *B. rubescens* (Table 4), due to the amount of biomass per area of the species (Table 3)

N was the nutrient that presented the highest stock in the foliar biomass of the highest AD species, varying from

0.24 to 4.17 kg ha^{-1}, with a mean of 1.09 kg ha^{-1} (Table 4).

The mean P stock in the foliar biomass of the regenerating species was lower than the other nutrients evaluated, ranging from 0.02 to 0.40 kg ha^{-1} (Table 4), mainly due to the low content in the leaves (Table 4). *B. rubescens* was the species that accumulated more P in the fragment due to high foliar biomass (Table 3). The species *I. capitata*, *C. densifolia* and *E. ovata* presented the lowest stock of P (Table 4).

The species *B. rubescens* and *T. mangle* showed the highest Ca stocks (1.37 and 0.87 kg ha^{-1}, respectively) and Mg (0.81 and 0.30 kg ha^{-1}, respectively), and the lowest stocks of Ca were found for *C. densifolia*, *I. capitata* and *E. ovata*. For Mg the smallest stocks were found in *C. densifolia* and *I. capitata*. *B. rubescens* was also the species with the highest stock of K and *C. densifolia* and *I. capitata* the smallest (Table 4).

The BUE of nutrients by natural regeneration species varied between nutrients and species. When nutrients were grouped in order of decreasing efficiency, the following sequence was obtained: P>Mg>K>Ca>N (Table 5).

Table 5. Biological utilization efficiency of nutrients of the species with highest absolute density (DA) of natural regeneration in a Lowlands Dense Ombrophilous forest fragment

Forest species	N	P	K	Ca	Mg
			kg kg^{-1}		
Brosimum rubescens	65.43 ±3.03a	687.08 ±80.65a	317.14 ±72.93a	201.39 ±30.32b	337.00 ±28.27c
Thyrsodium spruceanum	64.46 ±6.91a	731.51 ±73.49a	418.29 ±40.92a	158.04 ±49.14b	254.94 ±18.82c
Tovomita mangle	59.01 ±5.12a	754.16 ±70.90a	191.02 ±20.27b	84.01 ±16.96b	242.34 ±33.83c
Anaxagorea dolichocarpa	57.07 ±2.60a	408.32 ±19.09b	127.44 ±31.65b	70.10 ±4.93b	252.09 ±15.19c
Eschweilera ovata	53.79 ±5.24b	745.84 ±48.27a	240.30 ±54.37b	215.46 ±37.67b	299.69 ±60.90c
Protium arachouchini	64.58 ±6.33a	734.62 ±58.64a	212.98 ±47.69b	203.68 ±33.22b	606.95 ±85.71b
Caraipa densifolia	64.21 ±2.22a	670.49 ±86.02a	275.29 ±30.79b	282.90 ±36.16a	622.15 ±34.75b
Talisia retusa	66.71 ±4.03a	701.59 ±137.37a	220.10 ±82.55b	103.76 ±45.68b	235.66 ±30.33c
Inga capitata	42.32 ±5.15c	745.17 ±64.40a	233.29 ±9.91b	348.76 ±234.67a	457.25 ±27.13b
Protium heptaphyllum	66.79 ±9.57a	786.50 ±43.56a	352.11 ±137.12a	345.57 ±150.51a	1,099.95 ±400.72a
Mean	60.44	696.53	258.80	201.37	440.80
F$_{calculated}$	6.03**	6.18**	5.31**	3.50**	12.76**
CV (%)[1]	9.02	10.68	24.53	46.40	30.18

[1]Coefficient of variation = Standard deviation/Mean x 100.

Means followed by equal letters in the columns do not differ from each other at 5% probability level by the Scott-Knott's test.

** Significant at 1% probability by the F test. [ns]Not significant.

N was the nutrient that species used less efficiently (Table 5) due to the high content of this nutrient in the leaves of the species (Table 4). *I. capitata* presented low BUE of N, but, as it is a leguminous species, it may present good growth and development under high or low N availability in the soil (Table 1) due to its ability to obtain N through the symbiosis with bacteria that perform biological fixation of N.

P was the nutrient with the highest BUE by species, ranging from 408.32 to 786.50 kg kg^{-1} (Table 5). *A. dolichocarpa* presented lower BUE of P than the other species.

The species that showed best BUE of K were: *T. spruceanum, P. heptaphyllum,* and *B. rubescens* (Table 5). *C. densifolia, I. capitata*, and *P. heptaphyllum* were the species most efficients for Ca; *P. heptaphyllum* was the most efficient for Mg (Table 5). Specifically for this nutrient, the species presented a very wide variation of efficiency. *T. mangle* was more than 4.7 times less efficient than *P. heptaphyllum*.

4. Discussion

Studies of nutritional efficiency in natural regeneration species may be difficult because requiring the collection of all the foliar biomass (destructive sampling). In this study, the total foliar biomass (684.81 kg ha^{-1}) presented a value similar to that found by Socher (2004) (778.47 kg ha^{-1}) in a natural regeneration study (individuals greater than 1.30 m in height and DBH less than 15 cm) in a Mixed Alluvial Ombrophylous forest in Paraná For Barbosa (2012), methodological differences and other factors such as species specificity, stand's age, location climatic zone, soil fertility, and anthropogenic disturbances may influence the biomass of different forest fragments. Nutrient content data of this study were obtained in the warmer and wetter period of the year in which the nutrient absorption is higher than in the dry periods. With the nutrient content maximized, we can calculate the stock potential of the species and their biological utilization efficiency.

In this study the concentrations of N presented by the species were always higher than 15 mg kg^{-1}. According to Epstein and Bloom (2006), N content equal to or greater than 15 g kg^{-1} are considered normal for most plants. TOC levels were high in the surface layer (Table 1) showing that there was good N availability in the fragment and the species were benefited by this high availability with adequate N levels in the leaves. N is an essential nutrient at any stage of plant development and one of its functions is the formation of basic compounds in the plant's life cycle (amino acids, proteins, nucleic acids, among others) (Capaldi, 2002). Additionally, the amount of N in the plant influences the absorption of all nutrients, making it essential to maintain adequate concentrations of this nutrient for the plants (Epstein & Bloom, 2006). According to Bredemeier and Mundstock (2000), N availability is usually a limiting factor, influencing plant growth more than any other nutrient.

The highest N content was verified in the leguminous species. The legumes fix N through symbiosis with bacteria of several genera, being the most common with the genus *Rhizobium* sp. and this species of leguminous, that have ability to associate with microorganisms and allows the N of the air to be transformed into N compounds assimilated by plants, which can make the plant partially or totally independent of the external supply of this nutrient (Nogueira, O. Oliveira, C. Martins, & Bernardes, 2012).

The high N content in the leaves of *I. capitata* may be due the symbiosis of this leguminous with N-fixing bacteria. Although there is no record of specific symbiosis studies with the species *I. capitata* in tropical forests, the study performed by G. Almeida, Nascimento, A. Almeida, Cardoso, and Leal (2013) reported the potential for spontaneousnodulation or symbiotic relationship in different species of this genus.

Garay *et al*. (2003) evaluated the N content of two exotic forest species, one leguminous, *Acacia mangium* (Fabaceae), and another non-leguminous *Eucalyptus grandis* (Myrtaceae)*,* used in agroforestry and reforestation. The authors observed that the N content in the leaf of the leguminous *Acacia mangium* (15.6 g kg^{-1}) were almost twice the value found in *Eucalyptus grandis* (8.6 g kg^{-1}). The N content determined by Espig *et al*. (2008) in the native forest leguminous *Parkia pendula* and *Dialium guianense* in a Dense Ombrophilous forest fragment were of 22.96 and 20.83 g kg^{-1}, similar to those found in this study.

On mean, the Ca content this study were lower than those found by Espig *et al*. (2008), but the result was compatible with the expected content for the evaluated natural regeneration stratum, because high Ca content may be related to the low mobility of this element in plant tissues and to leaf longevity. Thus, it is expected that the older the leaf, the higher its Ca content (Boeger, Wisniewiski, Reissmann, 2005). The study by Espig *et al*. (2008) was carried out in plants in the adult stage, but is the main reference for Lowlands Dense Ombrophilous forest nutrition in northeast Brazil. Additionally, the soil Ca content of the fragment was very low (Table 1). The fragment is located in a region of high rainfall and cationic nutrients are very vulnerable to leaching.

Plants of natural regeneration are young and soil/plant/atmosphere water relations are dependent on adequate K content in the leaves of the species. According to Epstein and Bloom (2006), the opening and closing of the stomata depend on the flow of K, and its adequate content in young tissues is indispensable for obtaining the cells turgor.

Boeger *et al*. (2005) studied the nutrients in leaves of tree species in three successional stages of Dense Ombrophilous forest in southern Brazil, and obtained mean K contents similar to those obtained in this study, ranging from 3.2 to 5.2 g kg^{-1}. However, the K content of this study are below those found by Golley *et al*. (1978) and Espig *et al*. (2008), because the authors found mean contents of 14.3 and 12.23 g kg^{-1}, respectively. The low K content for the evaluated natural regeneration species in this study can be associated with the initial phase of their development and with the ecological group of these species (Table 2). According to S. R. Silva, Barros, Novais, and Pereira (2002), in the initial phase of growth, species of the final groups of the succession have low K requirement, or are efficient in using this nutrient in low availability conditions. The authors also affirm that the effects related to K are very small for the growth of some native forest species. The K levels of the soil of the fragment were also very low, as well as Ca (Table 1). The K because it is a monovalent cation is more vulnerable to leaching than Ca and the intensity of the rains in the region contributes to its leaching.

The soil Mg contents did not correlate with the leaf contents of the species, especially when comparing the Ca/Mg ratio of soil and species. In the soil, the Mg contents were higher than Ca (Table 1) and in plants the Ca/Mg ratio was on mean 2.4/1 (Table 4). The plants absorbed more Ca than Mg, even when there is little availability of this nutrient in the soil. Mg is a nutrient with fundamental importance in photosynthesis because it is the central atom of the chlorophyll molecule (Viera & Schumacher, 2009). Its deficiency compromises this molecule's synthesis, which affects the constitution and stability of thylakoids and results in poor chloroplast formation (Epstein & Bloom, 2006).

The mean P content in this study was higher than the found by Espig *et al*. (2008), which was of 0.95 g kg^{-1},

possibly because P has ample mobility within the plant and, therefore, tends to focus on younger organs (Viera & Schumacher, 2009). In addition, soil P levels were very low (Table 1) and should have limited their absorption. In tropical soils, P is the most limiting nutrient, as found by Ellsworth *et al*. (2017).

In general, tree species have a high P absorption capacity in their initial stage of growth. Their availability in the soil favors mainly the pioneers' initial growth (Flores-Aylas, Saggin-Júnior, Siqueira, & Davide, 2003). According to C. Souza, Tucci, J. F. Silva, and Ribeiro (2010), P limits growth and interferes with other nutrients absorption.

Species that develop under the same soil and climate conditions may differ in relation to their nutritional requirements, because they tend to be more or less demanding of a given nutrient. This situation occurs in forest fragments with high floristic diversity. According to Bündchen *et al*. (2013), this behavior emphasizes the importance of mixed stands to satisfy the nutritional demands of the species, because, for a better use of soil nutrients, a species that is more demanding of a nutrient should be closer to a less demanding species.

The heterogeneity, both of species and nutritional, is essential for forest self-sustainability, because the release of nutrients from the phytomass by decomposition and mineralization processes supplies a large part of the nutritional demand necessary for the growth and adequate development of forest species (Bündchen *et al*., 2013).

The species presented different levels of nutrients even when exploring the same soil conditions. Some factors may have contributed to this, such as ability to fix N_2 atmospheric, such as legumes; greater capacity of lateral and deep rooting, increasing its capacity of absorption of nutrients; formation of more or less dense crowns with greater or lesser biomass production. In this case, the nutrient can be diluted or concentrated in a larger or smaller biomass.

Nutrient stock was strongly influenced by the amount of biomass per area. According to Caldeira *et al*. (2002), the stock of nutrients is a consequence of their content and the production of biomass. In fact, the species studied showed differences in nutrient stocks.

The litter in forest fragments is important for the biogeochemical cycling process of nutrients, mainly for N, P, and Ca, because it is the main transfer route of these nutrients to the soil (Cole & Rapp, 1980). As shown, these three nutrients presented the highest stocks in the studied natural regeneration species. Therefore, as its leaves are incorporated into the litter and decomposed material, they'll contribute to the supply of these nutrients to the soil, and, consequently, will be available to plants in subsequent cycles

According to Espig *et al*. (2008), the nutritional stock evaluation is important in commercial plantations, where it predicts the nutrients export during the interest compartments removal, as wells as in natural forests, to predict the capacity of the species to supply nutrients and evaluate its contribution to the ecosystem balance, especially when dealing with low natural fertility areas.

The species of natural regeneration showed a wide variation in the BUE of nutrients. According to Bündchen *et al*. (2013), these differences evidenced the existence of different strategies for the acquisition and use of nutrients by tree species in the same establishment site.

The nutrient content found in the foliar biomass of the species in tropical soils is generally very low. S. R. Silva *et al*. (2002) stated that the decrease of the nutrient content in the soil solution causes to its less absorption by plants, resulting in an increase in BUE of nutrient. For example, P was the nutrient with the highest BUE by the species. This result showed that in weathered soils with low levels of available P, the species could develop properly, because they presented high BUE of this nutrient to produce biomass. In tropical forest soils, the P content are very low, as in this study and the phosphate nutrition of the forests is very dependent on the P cycling (C. Souza *et al*., 2010). As the cycling amount of P is also small (Bizuti, 2011), the high BUE of P of the species is fundamental for these species establishment, mainly in natural regeneration.

BUE indicates the ability of the species to grow in nutritionally restrictive environments (Barbosa, 2012). In this study, the highest BUE of N, P, K, Ca and Mg of the species can indicate them to be used in areas where there is a diagnosis of low levels of these nutrients, which contributes to reforestation projects, especially in tropical forests. In addition, the use of native species in these reforestation projects should be encouraged, especially when the nutritional demand of these species is known.

5. Conclusions

The BUE of nutrients by species varied according to the following decreasing order: P>Mg>K>Ca>N. In tropical soils of low natural fertility, the use of these species can be recommended in environmental reforestation projects;

In environment with restricted K availability, *T. spruceanum, P. heptaphyllum*, and *B. rubescens* can be recommended, as well as *P. heptaphyllum*, which can also be indicated to populate areas with low levels of Ca and Mg in the soil, common in tropical environments;

The difference of the nutritional demand between species may indicate for planting those with greater capacity to absorb and use nutrients, being most efficient in areas of low natural fertility soils of tropical forests.

Acknowledgments

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