

2
3 **Nitrogen Nutrition in Dry Tropical Forest at**
4 **Different Times of Regeneration**

5
6
7
8 **ABSTRACT**

9
10 The management of forest species requires the quantification of various nutrients flows in the ecosystem. Specifically for N this aspect is even more important because many species in dry forests, as Brazilian Caatinga, are legumes and in symbiotic association with diazotrophic bacteria, fix N₂ from the atmosphere. This study aimed to evaluate the N nutrition of forest species in Caatinga fragments with different regeneration times in the semi-arid region of Brazil. The study areas had different historical uses: the first one (53 ha) had no exploitation in the last 44 years; the second (32 ha) was in regeneration for 25 years; and the third (25 ha) had been in regeneration for five years. Four common tree species were evaluated in the three evaluated areas. Leaves were collected from each species to determine N contents. The legume species in the preserved area presented higher N content, decreasing with the regeneration time. The results showed a greater ecological balance in the most preserved sites, favoring the biological N fixation. N acquisition by legumes was not influenced by the disturbance of the site, suggesting the recommendation of these species as restorers of degraded areas.

11 *Keywords: Caatinga, forest nutrition, N cycling, leguminous forest species.*

12
13
14 **1. INTRODUCTION**

15
16 The mineralization of vegetal residues is the main route of nutrients supply to maintain soil fertility in forest ecosystems. Leaf compartment has already been shown to have greater contribution to the biomass supply in the soil and to the highest concentration of nutrients, particularly N, in different forest ecosystems [1-2].

17
18
19 In dry forest environments, such as Caatinga, a high number of deciduous species occurs, which confirms the high potential for nutrient cycling via foliar deposition. In general, the soils of the semi-arid region present low levels of N, and the mineralization of organic matter is the main source of this nutrient for the vegetation [3]. This condition is increased by vegetation degradation, in which most semi-arid environments are subject to [4].

20
21
22
23
24
25
26
27
28
29
30
31
32
33
34
Nutrient content in leaves of trees may be influenced by internal and external factors, as site conditions, physiological age of leaves, position of leaves in the canopy, time of year [5]. In soils deficient in some nutrients there is a greater demand for these, making the internal cycling inside the plants more active, as for N in soils from Brazilian semi-arid region.

Native leguminous species, adapted to the conditions of high temperatures and low water availability in semi-arid, may represent potential species to recover these adverse environments, mainly due to the biological N fixing capacity and high production of litter [6].

35
36
37
38
39
40
41
42
43
44
45
46
47
48
49
50
51
52
53
54
55
56
57
58
59

The management of forest species requires the quantification of various nutrients flows in the ecosystem. According to Bündchen [7], nutrient mobility and conservation in the forest ecosystem may be an important attribute and related, in part, to the ability of trees to occupy low fertility soils.

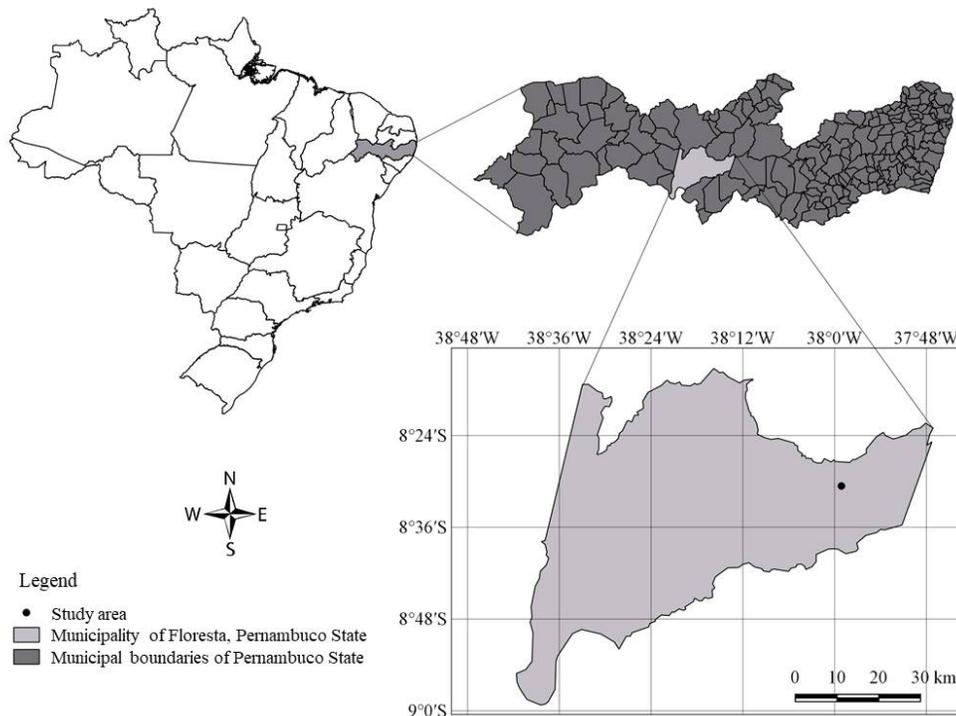
In the recovery of degraded soils with native forest species, the greatest difficulty has been the lack of studies involving the acquisition of nutrients and the nutritional requirements of these species, mainly for environments such as the semi-arid region [3]. Studies of this nature in disturbed ecosystems are important as a basis for understanding changes in the nutrient cycling process caused by anthropogenic interference and as a basis for assessing its recovery.

Thus, the objective of this study was to evaluate the N nutrition of forest species in three fragments of hyperxerophilic Caatinga with different times of regeneration after cutting for wood exploitation.

2. MATERIAL AND METHODS

2.1 Study area

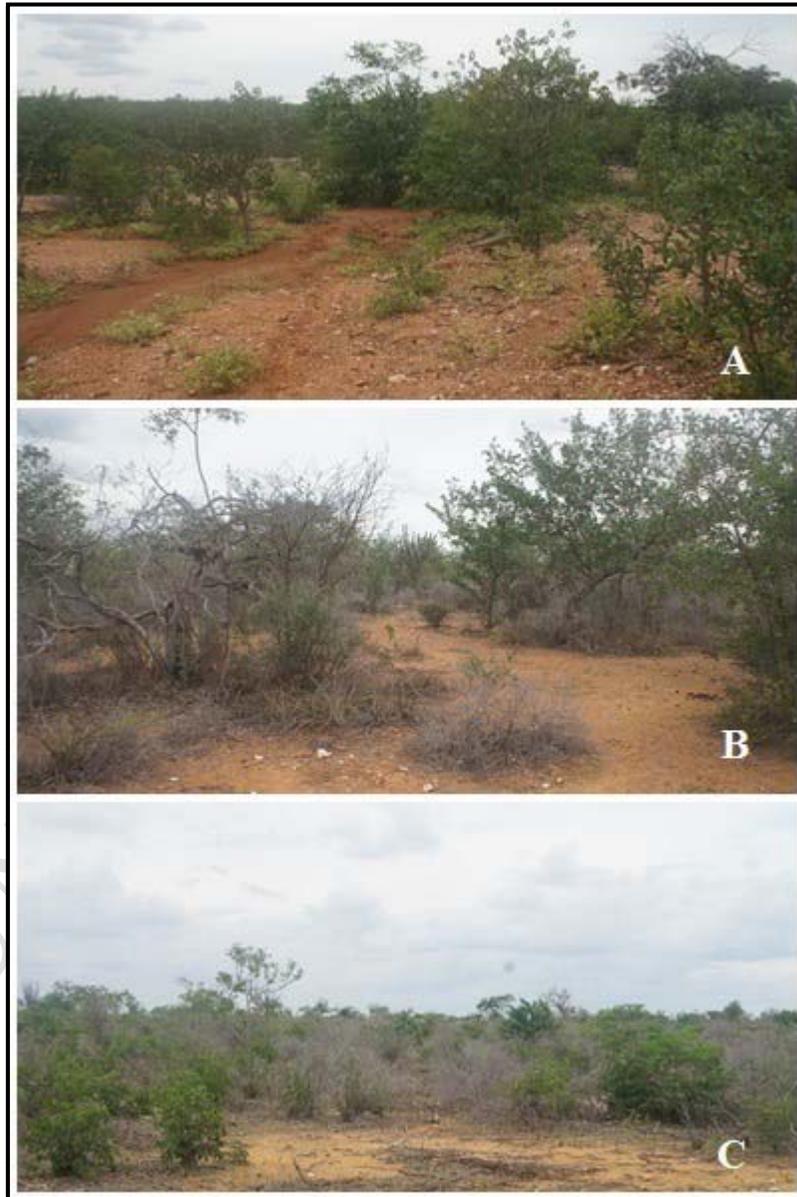
The study was carried out on Caatinga fragments located in the municipality of Floresta, Pernambuco, Brazil (Fig. 1).



60
61
62

Fig. 1. Geographic location of the study area, Brazil.

63 Three areas presenting different use histories and were selected to the study. The first one
64 (Preserved), with 53 ha, had not been explored in the last 44 years; the second place had
65 been in regeneration for 25 years after the cutting, and it is approximately 32 ha
66 (Regeneration 25 years); and the third site was cut five years ago (Regeneration 5 years)
67 and it is 25 ha extension (Fig. 2). Currently the regeneration areas have been managed for
68 the exploitation of firewood for charcoal and/or wood. The areas in regeneration for five and
69 25 years are distanced by about 200 m and are distant from the preserved area at about
70 2,000 m.



71
72
73
74
75
76
77
78
79
80
81
82
83
84
85
86
87
88
89
90
91
92
93
94
95
96
97
98
99
100
101
102
103
104
105
106
107
108
109
110
111
112
113
114

115 **Fig. 2. Fragments of Caatinga where the studies were carried out. Preserved Caatinga**
116 **area (A); Caatinga area in regeneration 25 years (B); and the regenerating Caatinga**
117 **Area 5 years (C).**

118
119 According to Köppen's classification, the climate in this municipality is BSh ' semi-arid, with
120 well-defined dry season, and the rainfall is concentrated mainly in the summer. The annual
121 average rainfall is 431.8 mm and the annual temperature is between 24 and 26 °C, providing
122 high evaporation rate and low relative humidity [8].

123 The vegetation is deciduous thorny woodland [9], characterized by shrub-tree vegetation.
124 The soil is classified as Chromic Luvisol, shallow and sandy to medium texture in the surface
125 [10].

126 Four tree species common in these three studied areas were evaluated. These species were
127 defined as having the highest importance value (IV) in the forest fragments in a floristic and
128 structural study carried out by Alves Júnior [11] (Table 1).

129
130 **Table 1. Forest species of natural regeneration of higher value importance (HVI) and**
131 **botanical family in fragments of the Caatinga, Brazil**
132

Species	Botanical family
<i>Poincianella bracteosa</i> (Tul.) L. P. Queiroz	<i>Caesalpinioideae</i>
<i>Mimosa ophtalmocentra</i> Mart. ex Benth	<i>Fabaceae Mimosoidae</i>
<i>Cnidoscolus quercifolius</i> Pohl	<i>Euphorbiaceae</i>
<i>Mimosa tenuiflora</i> (Willd.) Poir	<i>Fabaceae Mimosoideae</i>

133
134 **2.2 Species sampling and chemical analysis**
135

136 A sampling of leaves was performed to estimate the N content. Four individuals of each
137 species were sampled, having as criterion of selection the similarity in the size, vegetative
138 development and phytosanitary status of the individuals sampled. There were collected
139 randomly 25 newly mature leaves at the four cardinal points in a total of 100 leaves in each
140 plant (400 leaves by plot).

141
142 The sampled leaves were packed in paper bags and stored in coolers containing ice.
143 Subsequently, the samples were placed in an oven with forced air ventilation and maintained
144 at 65 ° C until constant weight. They were then crushed and stored in previously cleaned
145 and dried flasks for N analysis [12]. For N-total determination, the samples were digested in
146 sulfur solution and analyzed by the Kjeldahl method [13].
147

148 **2.3 Statistical procedures**
149

150 The statistical procedure for the study of N content data was the analysis of variance
151 (ANOVA) and averages comparison by Scott-Knott test ($p < 0.05$), when the main effects
152 and/or interactions were significant by F-test ($p < 0.05$).
153

154 The orthogonal contrast technique was used to analyze differences of the averages between
155 legume and non-legume species groups and for the groups of preserved versus in
156 regeneration areas. The difference between the means of contrast was evaluated by the t-
157 test ($p < 0.05$).

158
 159
 160
 161
 162
 163
 164
 165
 166
 167
 168
 169
 170

3. RESULTS AND DISCUSSION

N contents of the plant leaves in the hyperoxerophilic Caatinga fragment differed between regeneration times only for the species *Cnidoscolus quercifolius* and *Mimosa tenuiflora*. *Cnidoscolus quercifolius* had the highest N content in the area at 25 years of regeneration, while *Mimosa tenuiflora* showed the highest content in the area in 5 years under regeneration (Table 2).

Table 2. N content in leaves of forest species in Caatinga fragments at different times of regeneration, Pernambuco, Brazil

Forest species	Time of regeneration (years)			Average
	Preserved	5	25	
	g kg^{-1}			
<i>Cnidoscolus quercifolius</i>	29.96 aB	34.30 aB	43.65 aA	35.97
<i>Mimosa ophthalmocentra</i>	20.27 bA	17.36 bA	16.54 cA	18.05
<i>Mimosa tenuiflora</i>	24.82 aA	18.73 bB	27.34 bA	23.63
<i>Poincianella bracteosa</i>	17.87 bA	16.01 bA	17.22 cA	17.03
Average	23.23	21.60	26.18	
	F test			
Species				37.220*
Time				3.547*
Species x Time				2.823*
CV (%) ¹				20.86

171 ¹Coefficient of variation = standard deviation/Average x 100. Averages followed by the same
 172 letter, uppercase in the row and lowercase in the column, do not differ statistically by the
 173 Scott-Knott test (p<0.05). *Significant by F test (p<0.05).
 174

175 Although *Poincianella bracteosa* and *Mimosa ophthalmocentra* had no difference in N
 176 content in the area of higher equilibrium (preserved) or in regenerating environments, these
 177 species presented higher N levels in the preserved area. The acquisition of N by these
 178 legumes is higher because they are associated with N-fixing bacteria. On the other hand, the
 179 legume *Mimosa tenuiflora* showed to be more demanding in this nutrient presenting higher
 180 contents in the areas in regeneration (Table 2).
 181

182 *Cnidoscolus quercifolius* presented high levels of N in relation to legume species regardless
 183 of the regeneration time, which shows a high requirement in N, since this species is the only
 184 non-legume. All this strategy of survival can cause this species to regenerate much more

185 slowly than the others. This may indicate that regeneration of leguminous species is faster
186 and more balanced.

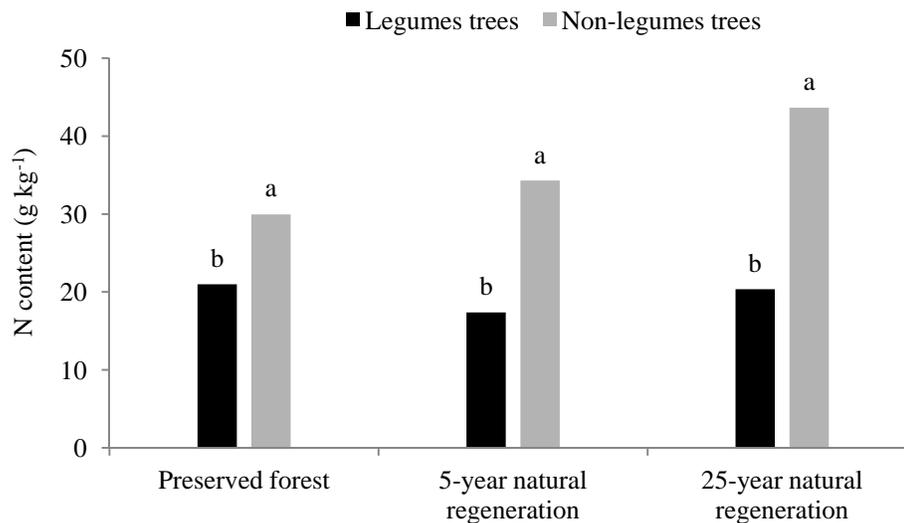
187

188 The results obtained showed that the evaluated species had N contents similar to those
189 found in studies in other forests in Brazil. Bündchen [7] obtained N levels between 23.85 and
190 35.56 g kg⁻¹ in a study with five species in subtropical forest in southern Brazil. In decidual
191 seasonal forest in Rio Grande do Sul, Vogel [14] found average N contents in the leaves of
192 24.20 g kg⁻¹. Mendes [15], evaluating ten native species in Central Amazonia observed N
193 contents in leaves varying between 15.70 and 22.40 g kg⁻¹. In Atlantic Forest, Cunha [2],
194 studying two fragments of Montana Forest in Rio de Janeiro observed average N contents in
195 leaves of 25.80 and 26.66 g kg⁻¹. In area of Caatinga in regeneration, Alves [3] obtained
196 average N contents in the leaves of five species ranging from 18.00 to 23.10 g kg⁻¹.

197

198 N contents in leaves of legume species were lower than that of non-legume species in all
199 evaluated environments (Figure 3). These results demonstrated the highest N requirement
200 for the only non-leguminous species (*Cnidoscolus quercifoilius*) and especially it was verified
201 that there was an increase of this species occurrence in less balanced areas, with shorter
202 times of regeneration.

203



204

205 Figure 3. Orthogonal contrast of N content in leaves of legumes versus non-legumes forest
206 species, separately for each time of regeneration in the Caatinga fragments, Pernambuco,
207 Brazil. Means followed by different lowercase letters at each regeneration time differ
208 statistically by the t test (p<0.05).

209

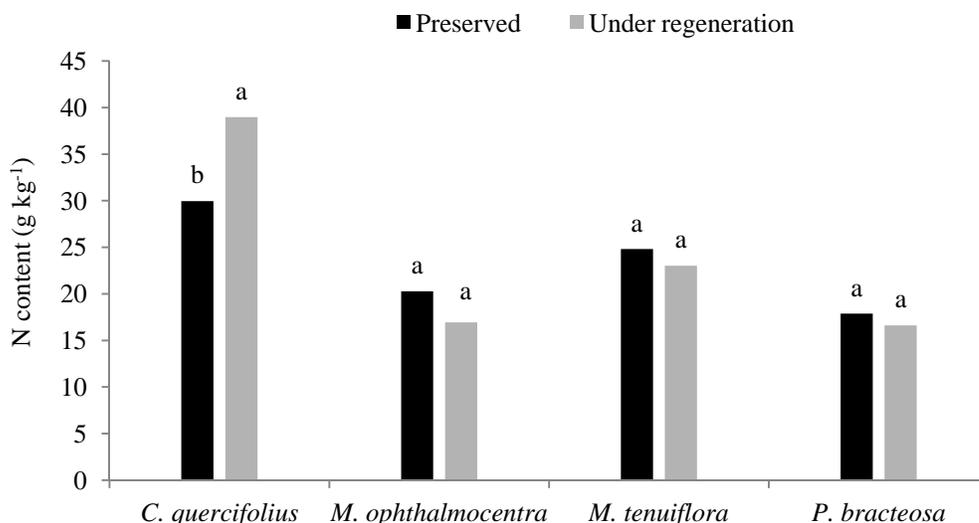
210

211 There was certain homogeneity of the average content of N in the leaves of legume species
212 in the different studied sites, varying between 17.36 and 20.98 g kg⁻¹ in the regenerating
213 area for five years and the preserved area, respectively (Figure 3). It was also verified that in
214 the preserved area, the leguminous species presented higher N content in the leaves,
215 decreasing with the reduction of the regeneration time of the areas. This may be related to
216 the greater ecological balance in the most preserved sites and with a longer regeneration
217 time, favoring the biological N fixation.

218

219 Concerning the contrast between the N levels in the leaves of the species and the
220 regenerated and preserved environments, it was verified that the legume species presented

221 similar results in these environments (Figure 4). It emphasizes the advantage of N biological
222 fixation for the recovery of degraded environments, legume species can be used in the
223 recovery of degraded areas, since the acquisition of N was not influenced by the disturbance
224 of the site.



225 Figure 4. Orthogonal contrast of the N content in leaves of forest species in the preserved
226 versus regenerated areas, separately for each forest species in the Caatinga fragments,
227 Pernambuco, Brazil. Means followed by different lowercase letters in each forest species
228 differ statistically by the t test ($p < 0.05$).
229
230

231 Regarding to *Cnidocolus quercifolius*, the levels of N in the regenerated and preserved
232 environments were inversely related to what occurred with the leguminous species, with
233 higher levels in the regeneration sites. This may indicate that in less balanced environments
234 *Cnidocolus quercifolius* tends to store more N and favor its internal cycling, because there
235 is higher demand for N by plants in these places, greater soil limitation and there is no
236 competitive advantage such as biological N fixation.
237

238 Moreover *Cnidocolus quercifolius* showed higher average N contents in the evaluated
239 environments than legume species.
240

241 4. CONCLUSION

242
243 The legume species in the preserved area showed higher N content decreasing with the
244 regeneration time, which it could be related to greater ecological balance in the most
245 preserved sites, favoring biological N₂ fixation. The acquisition of N by legumes was not
246 influenced by the disturbance of the site, suggesting the recommendation of these species
247 as restorers of degraded areas.
248

249 COMPETING INTERESTS

250
251 Authors have declared that no competing interests exist.
252

253 REFERENCES

254
255

- 256 1. Espig SA, Freire FJ, Marangon LC, Ferreira RLC, Freire MBGS, Espig DB. Composition
257 and efficiency of the biological use of nutrients in a fragment of the Atlantic Forest in
258 Pernambuco. Ci. Fl. 2008;18:309-316. Portuguese.
- 259 2. Cunha GM, Gama-Rodrigues AC, Gama-Rodrigues EF, Velloso ACX. Biomass and
260 carbon and nutrient stock in montane forests of the Atlantic Forest in the northern region
261 of the state of Rio de Janeiro. Rev. Bras. Ciênc. Solo. 2009;33:1175-1185.
262 <http://dx.doi.org/10.1590/S0100-06832009000500011>. Portuguese.
- 263 3. Alves AR, Ferreira RLC, Silva JAA, Dubeux Junior JCB, Osajima JA, Holanda AC.
264 Nutrient content in biomass and nutritional efficiency in Caatinga species. Ci. Fl.
265 2017;27:377-390. <http://dx.doi.org/10.5902/1980509827686>. Portuguese.
- 266 4. Holanda AC, Feliciano ALP, Freire FJ, Souza FQ, Freire SRD, Alves AR. Contribution of
267 litter and nutrients in an area of Caatinga. Ci. Fl. 2017;27:621-633.
268 <http://dx.doi.org/10.5902/1980509827747>. Portuguese.
- 269 5. Behling M, Neves JCL, Barros NF, Kishimoto CB, Smit L. Efficiency of nutrient
270 utilization for the formation of fine and medium roots in teak stands. Rev. Árvore.
271 2014;38:837-846. <http://dx.doi.org/10.1590/S0100-67622014000500008>. Portuguese.
- 272 6. Caldeira MVW, Sperandio HV, Delarmelina WM, Burak DL, Hunz SH. Biomass and
273 nutrient content in different above-ground compartments of *Mimosa velloziana* Mart and
274 *Tephrosia candida* D.C. Enflo. 2014;2:9-18. <http://dx.doi.org/10.5902/2316980X15225>.
275 Portuguese.
- 276 7. Bündchen M, Boeger MRT, Reissmann CB, Silva LSC. Nutritional status and nutrient
277 use efficiency in subtropical forest tree species in southern Brazil. Sci. For. 2013;41:227-
278 236. Portuguese.
- 279 8. Alvares CA, Stape JL, Sentelhas PC, Gonçalves JLM, Sparovek V. Köppen's climate
280 classification map for Brazil. Meteorol. Z. 2013;22:711-728. [http://doi.org/10.1127/0941-
281 2948/2013/0507](http://doi.org/10.1127/0941-2948/2013/0507).
- 282 9. Ferraz JSF, Ferreira RLC, Silva JAA, Meunier IMJ, Santos MVF. Structure of shrub-
283 arboreal component of vegetation in two areas of caatinga, in the municipality of forest,
284 Pernambuco. Rev. Árvore. 2014;38:1055-1064. [http://dx.doi.org/10.1590/S0100-
285 67622014000600010](http://dx.doi.org/10.1590/S0100-67622014000600010). Portuguese.
- 286 10. Santos HG, Jacomine PKT, Anjos LHC, Oliveira VA, Lumberras JF, Coelho MR. et al.
287 Brazilian system of soil classification. Rio de Janeiro: EMBRAPA; 2013. Portuguese.
- 288 11. Alves Júnior FT. Structure, biomass and volumetry of an area of Caatinga, Forest - PE.
289 [Thesis]. Recife: Universidade Federal Rural de Pernambuco, Brasil; 2010. Portuguese.
- 290 12. Silva FC. Manual of chemical analyzes of soils, plants and fertilizers. Brasília:
291 EMBRAPA; 2009. Portuguese.
- 292 13. Tedesco MJ. Analysis of soil, plants and other materials. Porto Alegre: Universidade
293 Federal do Rio Grande do Sul; 1995. Portuguese.
- 294 14. Vogel HLM, Schumacher MV, Trüby P. Biomass and macronutrients of a seasonal
295 deciduous forest in Itaara, RS, Brazil. Rev. Árvore. 2013;37:99-105.
296 <http://dx.doi.org/10.1590/S0100-67622013000100011>. Portuguese.
- 297 15. Mendes KR, Marenco RA, Magalhães NS. Growth and photosynthetic efficiency of
298 nitrogen and phosphorus use in Amazon forest species in the juvenile phase. Rev.
299 Árvore. 2013;37:707-716. <http://dx.doi.org/10.1590/S0100-67622013000400014>.
300 Portuguese.