



## **Biomass Production and Nutritional Characterization of *Eucalyptus benthamii* in the Pampa Biome, Brazil**

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### **Authors' contributions**

*This work was carried out in collaboration among all authors. Author CCG was responsible for the statistical analyzes and execution of the manuscript. Author DRM was responsible for the translation and adaptation of the manuscript according to the norms of the journal. Author MVS is advisor and contributed to the discussion of the data. Authors AAL and ACM helped in the discussion of the work.*

*Author HPS was responsible for making the experimental area available. All authors read and approved the final manuscript.*

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### **ABSTRACT**

The objective of this study was to evaluate the biomass production and to characterize a 7-year-old *Eucalyptus benthamii* stands in the Pampa-RS Biome. Initially, a sample inventory was performed for the dendrometric characterization of the stand. For the determination of biomass, nine trees were felled and fractionated in wood, bark, branch and leaves. Soil samples and plant tissues were collected and analyzed for nutritional characterization and determination of biological utilization coefficient (BUC). The average annual increment (AAI) with bark was 49.87 m<sup>3</sup> ha<sup>-1</sup>. The biomass production was 192 Mg ha<sup>-1</sup>, distributed in wood (81.2%)> branches (11%)> bark (6,5%)> leaves

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(1,3%). The leaves component presented the highest nutrient concentration and the wood the highest amounts of nutrients allocated in the biomass, except for Ca and Mg, observed in the bark. The highest BUC was observed in the wood. Mg was the nutrient that provided the best efficiency with a yield of 6,014 kg of wood per kg of Mg used, followed by S, P, Ca, K and N.

*Keywords: Forest soil; productivity; harvest; forest nutrition; nutrient cycling.*

## 1. INTRODUCTION

The natural population of *Eucalyptus benthamii* occurs in Australia, distributed along the eastern coast of New South Wales, southwest of the city of Sydney on alluvial plains on the banks of the Nepean River and its tributaries [1]. In this region, the species is distributed among only four populations, the largest of which consists of 6,550 trees and the other three with less than 340 trees. This condition places *E. benthamii* in extinction threat, with the following main factors: low seed viability due to the high degree of inbreeding and self-fertilization, low natural regeneration, competition with introduced species, changes in water regimes, fires, increase of urban areas and intense agricultural activity in the area of natural occurrence [2].

Regarding the silvicultural aspects, according to a study carried out in the Colombo-PR region, the species' high frost tolerance stands out, supporting absolute minimum temperatures of up to -10°C, besides presenting fast growth, uniform stem and high homogeneity of the plot [3]. These characteristics set *Eucalyptus benthamii*, as a good alternative for silvicultural use in cold climate regions, especially where there is frequent and severe frost occurrence, as in southern Brazil [4].

The evaluation of the productivity, biomass and nutrient production at the end of the rotation in a forest plantation, can help in the decision making of the forester concerning the choice of the species to be implanted and the nutritional replacement of the new production cycle. According to Viera et al. [5], the choice of efficient genotypes to absorb and use nutrients must be performed in order to improve applied fertilization.

In addition, the applied harvest intensity, with removal of one or more components of the tree, directly affects the export of nutrients from the forest site. In order to maintain the soil productive capacity [5], the nutritional replacement cost would be increased from corrective fertilization

and maintenance. According to Achat et al. [6], the removal of the residues causes a decrease in the biological activity and an increase in soil compaction, and as a result there is a decrease in growth.

In this sense, the objective of this work was to evaluate the biomass and nutrient production and determine the nutrient utilization efficiency of an experimental plantation of *Eucalyptus benthamii* Maiden & Cabbage in the Pampa Biome of Rio Grande do Sul, Brazil.

## 2. MATERIALS AND METHODS

### 2.1 Characterization of the Experimental Area

The experiment was carried out in an area of 10 hectares of *E. benthamii*, seven years old, in the municipality of Alegrete-RS, with central geographical coordinates: 55°32'53" West longitude and 29°47'60" South latitude.

The climate of the region is classified as humid sub-temperate, with frequent frosts from May to August, and intense heat in summer, mainly in the months of January and February, with the average temperature of the month being > 22°C and average annual temperature > 18°C. Annual precipitation presents rainfall indexes ranging from 1,250 to 1,500 mm [7]. The soil of the experimental area is classified as typical Distrophic Red Argisol [8].

The planting of the seedlings was done manually and without irrigation, using seminal seedlings, with initial density of 1428 plants ha<sup>-1</sup> (3.5 m x 2.0 m). Subsoiling was performed 30 days before planting, using a subsoiler with three stems, incorporating 300 kg ha<sup>-1</sup> of reactive natural phosphate (GAFSA, 12% P<sub>2</sub>O<sub>5</sub> soluble in citric acid) followed by light harrowing.

Fertilization was carried out 15 days after planting, using the formula N-P<sub>2</sub>O<sub>5</sub>-K<sub>2</sub>O from 06-30-06 + 0.6% B, 110 g plant<sup>-1</sup>, divided into two

sub-doses of 55 g incorporated at 15 cm distance on each side of the seedling. The second fertilization was carried out at 90 days post-planting, using fertilizer with formulation N-P<sub>2</sub>O<sub>5</sub>-K<sub>2</sub>O from 20-05-20 + 0.2% B + 0.4% Zn, 122 g plant<sup>-1</sup>, applied manually for the canopy projection. The third fertilization, at 270 days, was used the formula N-P-K<sub>2</sub>O of 22-00-18 + 1% S + 0.3% B, 122 g plant<sup>-1</sup> applied mechanically in the interlining. At no time was liming performed.

## 2.2 Experimental Design and Data Collection

Through the forest inventory the growth variables were obtained. Four plots (35 m x 20 m) were randomly distributed, all diameters at breast height were measured (DBH) with diametric tape and all tree heights (m) with Vertex hypsometer. After the measurements the trees were distributed in three diameter classes, the first class being from 10 to 16 cm, the second from 16.1 to 22 cm and the third class from 22.1 to 28 cm.

For the determination of above-ground biomass (Mg ha<sup>-1</sup>), three trees were selected by diameter class, with a tree at the lower limit, a tree at the central limit and a tree at the upper limit of each class, totaling nine individuals felled. The selected trees were sectioned at ground level and cubed by Smalian methodology, as described by Finger [9]. After the canopy, each felled tree was fractionated in the components: leaves, branches, bark and wood. Each component had its biomass measured in the field by weighing with hook scale, with accuracy of 50 g.

A sample of leaves and branches was collected per evaluated tree. For wood and bark, three samples per tree distributed along the commercial shaft with a minimum diameter of 8 cm were collected, in the median positions of the sections resulting from the division into three equal parts of the same. All the samples were weighed in the field, properly packed, identified and sent for analysis in laboratory.

The estimated tree biomass per hectare was determined by regression analysis applied to the inventory data and extrapolation based on the sample unit area. The amount of macronutrients (kg ha<sup>-1</sup>) and micronutrients (g ha<sup>-1</sup>), allocated into tree components, was obtained by multiplying the content of each nutrient by the biomass value.

For soil chemical analysis and density, samples were collected at depths of 0-20, 20-40 and 40-100 cm. Density determination followed the methodology proposed by Embrapa [10]. Plant tissue and soil analyzes were performed following the methodology described by Tedesco et al. [11] and Miyazawa et al. [12].

## 2.3 Statistics and Data Analysis

The Berkhout, Schumacher-Hall, Hohenadl-Kreen and Spurr models were used to determine the equations for height and volume estimation. The modeling of the equations for the individual biomass of the trees and their respective components was processed using the program "proc stepwise" - "forward" option of the statistical program SAS [13].

The biomass of each component and its arithmetic and logarithmic variants (natural logarithm) were considered as dependent variables. The independent variables were DBH (cm), height (m) and volume (m<sup>3</sup> tree<sup>-1</sup>) and their respective arithmetic and logarithmic variants (natural logarithm). The quality of the adjustment and selection of the equations considered as the main statistics, the highest adjusted coefficient of determination and the lowest relative standard error of the estimate (Syx%) were determined.

The contrast of the averages of the chemical and physical attributes of the soil between the different depths and, for nutrient contents in the biomass components (leaves, branches, bark and wood) was evaluated by Tukey test at the level of 5% probability of error. A completely randomized design was used for the statistical analysis, with the treatments being the different soil depths and the biomass components above the soil.

The biological utilization coefficient (BUC) was evaluated. The BUC can be described as the biomass nutrient conversion rate, obtained through the ratio between the biomass and the nutrient quantity, both with the same unit [14].

## 3. RESULTS AND DISCUSSION

### 3.1 Soil Fertility

According to the Soil Chemistry and Fertility Commission - RS / SC [15], the soil of the experimental area presents: textural class 4 (clay content ≤ 20%); low organic matter content (≤ 2.5); very low pH in water (≤ 5.0); low

exchangeable Ca content ( $<2.0 \text{ cmolc dm}^{-3}$ ); low exchangeable Mg content ( $\leq 0.5 \text{ cmolc dm}^{-3}$ ); high S content ( $> 5 \text{ mg dm}^{-3}$ ); the very low available P content ( $\leq 7.0 \text{ mg dm}^{-3}$ ); tvery low exchangeable K content ( $\leq 15 \text{ mg dm}^{-3}$ ); high content of B, Cu and Zn ( $> 0.3 > 0.4$  and  $> 0.5 \text{ mg dm}^{-3}$  respectively), very high saturation by Al ( $> 40\%$ ) and high saturation by bases ( $<45\%$ ).

Regarding the physical attributes, Lemos and Santos [16] classify the soil as sand-free surface texture and sandy loam clay texture in depth. According to Reinert and Reichert [17], the bulk density found is considered adequate for most crops (Table 1).

Despite the low fertility observed in the soil of the experimental area, the genotype presented good growth behavior, with an annual average increment with bark of  $49.87 \text{ m}^3 \text{ ha}^{-1}$  and a total production of commercial wood with bark of  $349.09 \text{ m}^3 \text{ ha}^{-1}$ . Among the models tested, for the estimation of height and volume, the best adjustments were provided by the Hohenald-Kreene and Schumacher-Hall models, respectively (Table 2).

The values of DBH, H, B, AAI and V verified are similar to those found by Benin [18], studying 6 years old *E. benthamii*, planted at different spacing, in Guarapuava-PR. However, it differed from Mendoza [19] which observed an average annual increment of  $34 \text{ m}^3 \text{ ha}^{-1}$  in a settlement of 7 years old *E. benthamii* with 85% of survival in northern Argentina.

The modeling of above-ground biomass in a 10 years old clone of *Eucalyptus saligna* stand was performed by Momolli et al. [20]. The authors found 89; 5.9; 3.2 and 1.8% of the biomass in the wood, bark, branch and leaf respectively. The average annual increment of bark was  $54.6 \text{ m}^3 \text{ ha}^{-1}$  and the total biomass above-ground was  $269 \text{ Mg ha}^{-1}$ . The highest percentages of wood occur due to the maturity of the stand, in addition to being a clone with genetic improvement.

Barros and Comerford [21] explain that the great variation in productivity of eucalyptus plantations at different regions is mainly associated to the different types of soils, presenting total and available nutrients contents in a very wide range. Considering this condition, Guimarães [22] adds

that the forester should intervene in the management of the site and, consequently, increase the gains in production and reduce operating costs.

The models selected for biomass estimation presented good predictive capacity and significance, evidenced by the equation adjustment statistics. The biomass production above-ground was  $192.0 \text{ Mg ha}^{-1}$ , distributed in wood (81.2%)> branches (11%)> bark (6.5%)> leaves (1.3 %) (Table 3).

Hernández et al. [23], evaluating 9 years old *E. dunnii*, in Uruguay, verified that the biomass production of wood was  $144 \text{ Mg ha}^{-1}$  and in the branches  $22 \text{ Mg ha}^{-1}$ , similar to this study, but for biomass of the bark ( $29 \text{ Mg ha}^{-1}$ ) and leaves ( $13 \text{ Mg ha}^{-1}$ ) the values observed were higher. Viera et al. [14] studying 10 years old hybrids of *Eucalyptus urophylla* x *E. globulus*, in Rio Grande do Sul, also verified a similar biomass production ( $167.10 \text{ Mg ha}^{-1}$ ). Guimarães et al. [24] studying a 4 years old *E. dunnii*, in the same region of this study, found an above-ground production of  $104.5 \text{ Mg ha}^{-1}$ , with  $76.7 \text{ Mg ha}^{-1}$  for wood biomass, differing from that observed in this study.

In relation to the relative partition, Schumacher et al. [25] evaluating *Eucalyptus* spp. stands, at different ages, observed that at 2 years old, 47% of the biomass was allocated to the wood, while at 8 years old, the proportion of biomass in the wood increased to 74.4%, with the reduction of the relative biomass of the other components, corroborating the trend found in this work. For Schumacher [26], the difference in biomass allocation in the tree components is very dynamic due to carbohydrate distribution, resulting from the photosynthesis, besides edaphoclimatic factors, species density of planting and age of the stand.

The highest nutrient contents of the biomass components were observed in the leaf, except of Ca and Mg, with higher values in the bark ( $p < 0.05$ ). The largest stocks of nutrients in above-ground biomass were observed in the wood, except for Ca and Mg, which were more accumulated in the bark. The highest nutrient utilization efficiency was verified in wood, which presented the highest values for the biological utilization coefficient, except for the Zn that was in the leaves component (Table 4).

**Table 1. Physical and chemical attributes of the soil in a 7 years old *Eucalyptus benthamii* stand, in Pampa Biome**

Depth	Density	<sup>(1)</sup> CS	<sup>(2)</sup> FS	Silt	Clay	OM	pH	CTC <sub>efet.</sub>	Ca
cm	g cm <sup>3</sup>			%			(H <sub>2</sub> O)	cmol <sub>c</sub> dm <sup>-3</sup>	
0-20	1.62a	75a	6a	3a	16a	0.7a	4.2a	2.3a	0.4b
20-40	1.57a	74a	4a	5a	17a	0.7a	4.3a	2.3a	0.5b
40-100	1.48a	69a	6a	5a	20a	0.6a	4.4a	2.7a	0.9a
Depth	Mg	P	K	S	B	Cu	Zn	V	m
cm	cmol <sub>c</sub> dm <sup>-3</sup>			mg dm <sup>-3</sup>			%		
0-20	0.1b	8.3a	26.9a	5.6a	0.6a	3.5a	0.3a	5.7b	75.2a
20-40	0.1b	4.0b	18.4ab	6.2a	0.6a	3.9a	0.1a	7.2b	70.8a
40-100	0.3a	3.9b	15.9b	4.5a	0.5a	3.5a	0.1a	13.1a	54.7b

(<sup>1</sup>)CS = Coarse Sand; (<sup>2</sup>)FS = Fine Sand; OM = Organic Matter; Equally vertical letters do not differ statistically between the attributes in the 0-20, 20-40 and 40-100 cm layers, respectively, at the 0.05 level of significance, by the Tukey test

**Table 2. Dendrometric characteristics of 7 years old *Eucalyptus benthamii* stands in Pampa Biome**

N	DBH			H			G			AAI <sub>wb</sub>	V <sub>wb</sub>	AAI <sub>b</sub>	V <sub>b</sub>
	Mean	$\sigma$	CV	Mean	$\sigma$	CV	Mean	$\sigma$	CV				
922	19.38	4.92	25.38	25.67	3.94	15.35	24.60	0.01	46.17	44.46	311.12	49.87	349.09
Model										Regression adjustment statistics			
										Prob. > F	R <sup>2</sup> adj.	Syx(%)	
H = -22.868938 + 3.305093.DBH - 0.0577850.DBH <sup>2</sup>										0.0001	0.91	10.51	
Log V <sub>b</sub> = -3.634929 + 1.5414769.logDBH + 0.894537.LogH										0.0001	0.99	7.21	
Log V <sub>wb</sub> = -3.688916 + 1.4279525.logDBH + 0.995249.LogH										0.0001	0.99	8.14	

N = Number of trees per hectare, DBH = diameter at breast height in cm, H = Total height in m, B = basal area in m<sup>2</sup> ha<sup>-1</sup>, CV = coefficient of variation; AAI = Annual average increment m<sup>3</sup> ha<sup>-1</sup>; V = Volume m<sup>3</sup>; wb = without bark; b = with bark

**Table 3. Quantity of biomass and models by components of 7 years old *Eucalyptus benthamii* stand, in the Pampa Biome**

<sup>(1)</sup> Mg ha <sup>-1</sup>	Model	R <sup>2</sup> adj.	Syx(%)
L	2.5 y = -0.192074 <sup>ns</sup> + 0.000273411** .d <sup>2</sup> h	0.93	20.26
Br	21.2 y = 13.87236** + 10.23822* .hv - 0.00049041** .d <sup>2</sup> h <sup>2</sup> + 0.00691** .d <sup>3</sup>	0.94	11.52
Ba	12.6 y = -1.80420 <sup>ns</sup> + 74.59571** .v - 1.162225* hv	0.96	6.61
W	156.0 y = -22.33094* + 869.02702** .v - 0.00034689 .d <sup>2</sup> h <sup>2</sup>	0.99	4.76
T	192.0 y = -44.85056** + 1348.85096** .v - 0.00071892** .d <sup>2</sup> h <sup>2</sup> + 122064 <sup>ns</sup> . 1/h <sup>3</sup>	0.99	3.58
C	23.7 y = 13.96618** + 10.78749** hv - 0.00050939** .d <sup>2</sup> h <sup>2</sup> + 0.00727** .d <sup>3</sup>	0.97	9.13
WBa	18.3 y = -23.88367* + 939.37265** .v - 0.00037597 <sup>ns</sup> .d <sup>2</sup> h <sup>2</sup>	0.99	4.48

Were: L = leaf; Br = branch; Ba = bark; W = wood; T = total; C = canopy; WBa = wood + bark; d = diameter at breast high; h = high; v = volum (m<sup>3</sup>); R<sup>2</sup> adj. = adjusted coefficient of determination; Syx (%) = standard error of estimate; ns = no-significant; \* Significant at 5% probability level of error; \*\* Significant at 1% probability of error

The magnitude of nutrient concentration, in decreasing order, was as follows: leaves > bark > branches > wood. The higher levels of nutrients observed in the leaf, as well as the difference in concentration between the biomass components, can be explained by the tendency of most nutrients to concentrate in the new tissues of the plant, where the main metabolic processes

occur. The interaction of these processes is intrinsically related to biochemical cycling, where with age, nutrients from senescent tissues tend to move to regions with higher metabolic activity, and biochemical cycling is more important for the maintenance of nutrients with high mobility (N, P, K and Mg), and lower for low mobility nutrients (Ca, S) and micronutrients [27,28,29].

**Table 4. Concentrations, nutrient amounts and biological utilization coefficient (BUC) of the above-ground biomass components of 7 years old *Eucalyptus benthamii* stand, in Pampa Biome**

Biomass	N	P	K	Ca	Mg	S	B	Cu	Mn	Zn
	g kg <sup>-1</sup> Concentration					mg kg <sup>-1</sup>				
L	31.0a	2.1a	12.7a	6.2b	2.3a	1.5a	32.0a	11.4a	1058.5a	16.8a
Br	6.5b	0.8b	4.8b	9.7b	1.9a	0.5b	10.9b	8.4ab	479.7b	22.5a
Ba	8.0b	1.3b	6.4b	19.7a	3.6a	0.6b	15.5b	5.1b	825.6ab	22.8a
W	1.9c	0.4c	1.5c	0.8c	0.1b	0.3b	4.5c	2.2c	77.2c	29.8a
	kg ha <sup>-1</sup> Amount					g ha <sup>-1</sup>				
L	77.5 (12.3)	5.0 (5.0)	32.1 (6.7)	12.3 (2.2)	5.0 (4.6)	3.6 (5.1)	80.5 (6.7)	27.1 (4.5)	2256.0 (6.4)	39.5 (0.6)
Br	137.0 (21.8)	16.7 (16.6)	106.9 (22.4)	172.5 (31.0)	37.0 (33.9)	9.2 (13.2)	237.5 (19.8)	175.4 (29.2)	9385.3 (26.6)	440.0 (7.1)
Ba	82.3 (13.1)	15.1 (15.0)	80.0 (16.8)	248.1 (44.5)	41.4 (37.9)	6.0 (8.5)	164.0 (13.7)	54.6 (9.1)	10825.1 (30.7)	222.7 (3.6)
W	332.7 (52.8)	63.6 (63.4)	257.6 (54.1)	124.1 (22.3)	25.9 (23.6)	51.4 (73.2)	692.8 (59.8)	374.6 (57.2)	12798.3 (36.3)	5462.0 (88.7)
	BUC									
L	32	498	78	204	507	706	31181	92765	1113	63510
Br	155	1265	198	123	573	2297	89163	120765	2257	48131
Ba	152	831	157	51	303	2088	76543	229874	1159	56359
W	468	2450	605	1256	6014	3030	224817	415795	12170	28517

Were: L = leaf; Br = branch; Ba = bark; W = wood. Vertical letters do not differ statistically between the nutrient contents in the biomass components, at 0.05 level of significance, by the Tukey test. Values in parentheses refer to the relative partition (%) of each nutrient per component in relation to the total amount.

Considering the nutrient utilization efficiency of wood, it was verified that Mg is the macronutrient least required to produce biomass, followed by S, P, Ca, K and N. Several authors have observed a higher conversion of nutrients to wood than those observed in this study. Silva et al. [30] evaluating five different eucalyptus species at 10 years of age, in Itirapina-SP, verified that P provided the highest biological utilization coefficient (BUC) of nutrients, especially *E. grandis* with 43441 for P in wood, but for Mg efficiency was lower among all genotypes. This behavior was also verified by Viera et al. [14] studying the hybrid *Eucalyptus urophylla* x *E. globulus*, with a BUC of 13285 for P in the wood, followed by S, Mg, Ca, K and N; and by Guimarães [22] studying the clonal hybrid *Eucalyptus urophylla* x *E. grandis*, *E. grandis* and *E. dunnii*, at 4 years old in Alegrete-RS, who verified in the *E. urograndis* a BUC of 17060 for P in the wood, followed by Mg, Ca, S, K and N. In another study carried out with eight clonal *E. urograndis* hybrids, at 9 years of age in Aracruz-ES, Neves [31] verified that S was the nutrient that presented the highest conversion in

wood (average BUC of 8500), followed by P, Mg, K, N and Ca; which was also observed by Beulch [32], studying 4 years old clones of *E. saligna*, in São Francisco de Assis-RS, who verified a BUC of 11688 for S in wood, followed by P, Mg, Ca, S, N and K.

Silva et al. [30] argue that the high efficiency presented by a species in the use of nutrients indicates a lower nutritional requirement, which can be used as an indicator for the selection of species that can be cultivated mainly in soils with low natural fertility. Santana et al. [33] adds that the use of genetic material with efficiency compatible with soil fertility can contribute to the maintenance of the productive capacity of the site and, consequently, use smaller amounts of fertilizers.

According to our study, there is a great potential for growth and production of the *Eucalyptus benthamii* genotype. The species showed to be less efficient in nutrient utilization, when compared to several seminal and clonal materials, cultivated on a large scale in Brazil.

However, considering its good adaptability to cold climate regions such as the Pampa Biome in the state of Rio Grande do Sul, further studies should be devoted to the genetic improvement of *Eucalyptus benthamii* through hybridization with species that present a better efficiency of nutrient utilization in this region and subsequent cloning of the best individuals for commercial cultivation.

#### 4. CONCLUSION

Despite the low natural fertility of the soil in the experimental area, *Eucalyptus benthamii* presented productivity similar to the other eucalyptus species cultivated on commercial scale in Brazil. The above-ground biomass is predominantly allocated to the wood (81.2%), followed by branches (11%), bark (6.5%) and leaves (1.3%).

The leaves present the highest levels of nutrients, except Ca and Mg (bark) and Zn (wood). The largest amounts of nutrients are allocated to the wood, except for Ca and Mg (bark). The wood presents the highest efficiency of nutrient utilization, except of Zn.

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#### COMPETING INTERESTS

Authors have declared that no competing interests exist

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