

DETERMINATION OF THE EFFECTIVE DOSE OF PHOSPHORUS IN LEMON EUCALYPTUS PLANTS (*Corymbia citriodora*)

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ABSTRACT

Phosphorus (P) is one of the most limiting nutrients for plant species and its intensive use can exhaust world reserves. In general, forest production is unviable without the availability of P. The objective of the study was to determine the effect of phosphate fertilization on plant growth of four families of lemon eucalyptus (*Corymbia citriodora*). Plants produced from seeds of four seed trees were used, which were subjected to seven doses of phosphorus: 0, 25, 50, 75, 100, 125 and 150 mg P dm⁻³. The experiment was carried out in a 7 × 4 factorial arrangement, with four replicates and using containers with 5 dm³ of a Red Eutrophic Latosol type soil. The response variables measured were monthly plant height, plant collar diameter at 90 d, dry biomass weight of leaves, roots, stems and branches, and phosphorus use efficiency of leaves. The variations of the characteristics evaluated as a function of phosphorus concentration were analysed by means of polynomial regression equations. Phosphate fertilization is necessary for the growth and development of *C. citriodora* plants. Plants of the four *C. citriodora* seed trees from different provenances responded differentially to phosphorus doses. It was found that the optimum amount of phosphorus ranges between 30 and 100 mg dm⁻³ according to the characteristics evaluated and the seed tree.

Keywords: seed trees, forest fertilization, provenances, phosphorus doses, *Corymbia citriodora*.

INTRODUCTION

Lemon eucalyptus (*Corymbia citriodora* Hill & Johnson) is a medium to large species, which can reach 50 m in height and 1.2 m in trunk diameter, with excellent trunk shape and sparse foliage. This species has good silvicultural characteristics, one of which is the quality of the wood it produces (Morais *et al.*, 2010).

Studies of nutrient use efficiency among clones of forest species carried out in greenhouses are useful for genetic improvement programs that seek early selection of these species, since the responses are obtained in short periods of time. Determining the adequate phosphorus dose is fundamental for the optimization of inputs such as chemical fertilizers and thus minimize losses.

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The use of synthetic fertilizers in forestry production is increasingly intense, especially in developing countries, and this encourages studies to minimize leaching losses and water contamination. In Latin America, the main fertilizer consuming countries of commercial groups are Brazil, Argentina, Mexico, and Colombia, which together with the other countries in this region are highly dependent on N, P₂O₅ and K₂O imports (FAO, 2015). Therefore, a more appropriate use of different fertilizer sources is necessary to promote sustainability.

In the early stages of development, the supply of phosphate fertilizers to *C. citriodora* seedlings allows the identification of the potential in the efficiency of use of this element. For example, Lima *et al.* (2005) identified differences in P utilization capacity among eucalyptus clones. One of the main effects of P deficiency in plants is expressed in a decrease in leaf growth, as well as in the number of leaves (Dussan *et al.*, 2016). The quality of forest seedlings can be evaluated by parameters based on phenotypic aspects, called morphological aspects such as height of the above ground part, diameter of stems, number of leaves and dry biomass weight of the above ground part and roots of the plants (Oliveira *et al.*, 2017). Thus, these parameters are fundamental for forest nutrition.

In soybean (*Glycine max*), phosphate fertilization increased productivity, the weight of one thousand seeds and pod and grain production per plant (Filho *et al.*, 2013). P is one of the most demanded macronutrients by plants and must be available at critical stages such as early seedling development after seed germination (Oliveira *et al.*, 2017). The hypothesis proposed in this study is that P plays a determining role in the initial development of lemon eucalyptus seedlings. The objective of this research was to determine the effect of different doses of phosphate fertilizers on the initial growth of *Corymbia citriodora* plants.

MATERIALS AND METHODS

Seed tree location areas

Seeds were used from lemon eucalyptus (*Corymbia citriodora*) trees from different seed production areas located at the Faculty of Agrarian and Veterinary Sciences of the Paulista State University (UNESP/FCAV), Jaboticabal Campus, São Paulo (tree number 4); at the São Paulo State Forestry Institute (tree number 7); at the Guarani Orchard (HG), Pradópolis, São Paulo (tree number 21); and at Borebi, São Paulo (tree number 30), in Brazil.

Phosphorus concentration and content analysis in leaves

Chemical analyses to determine the concentration of phosphorus in the leaves of the plants were carried out at the Horticultural Products Post-Harvest Laboratory of the Plant Production Department of UNESP. For this purpose, all the leaves of each tree were washed with 2 % neutral detergent, then dried with paper towel. Once dried and at constant weight, the leaves were crushed individually per tree (preserving the identity), using a steel grinder. The P concentration and the dry biomass obtained were used to estimate the P content.

Production of *C. citriodora* plants in nurseries

The plants were produced from seeds, which were placed in rigid plastic conical tubes with a capacity of 55 cm³ and Plantmax[®] Florestas composed of pine bark (*Pinus* spp.) and vermiculite (50:50, v:v) was used as substrate. These plants were produced in a nursery covered by a screen with 30% solar reduction and intermittent micro-sprinkler irrigation with 8 min duration and 45 min intervals with automatic control. When they reached the mean height of 10 cm and the age of 60 d after sowing, the plants were transplanted into 10 L plastic containers (capacity of 7 dm³), for the experiments with different doses of phosphorus.

The experiment was carried out inside a greenhouse with dimensions of 30 m long, 5 m wide and 4.5 m high, with a roof covered with plastic and side walls with anti-aphid netting.

Statistical design

The experiment was conducted in a randomized block design in a 7 × 4 factorial arrangement, where the study factor “P dose” had seven levels: 0, 25, 50, 75, 100, 125 and 150 mg P dm⁻³ and the factor “seed trees” four levels: Tree 4, Tree 7, Tree 21, and Tree 30. Each of the 28 treatments had four replications, for a total of 112 plots. Each plot consisted of a pot containing two plants as experimental unit, with a total of 224 plants. Triple superphosphate powder recommended for plants in seed production areas was used as a P source.

Soil collection and analysis

A Red Eutrophic Latosol type soil was used, collected at a depth of 0 to 20 cm, in the Hacienda Guatapar, sector A, under a high voltage electrical network with the following geographic coordinates: 21° 28' 19.17" S and 47° 58' 26.18" W. This location belongs to International Paper in the District of Luiz Antonio, SP, Brazil.

The soil was dried under shade, de-throned, sieved using a 4 mm opening sieve and then homogenized. A composite soil sample was removed for chemical analysis according to the methods proposed by Raij *et al.* (2001). The soil was analysed at the Soil Fertility Laboratory of the Soil and Fertilizer Department of FCAV-UNESP, Jaboticabal-SP, Brazil. The results obtained were phosphorus (P) extracted with resin: 4 mg dm⁻³; organic matter (MO): 19 g dm⁻³; pH in CaCl₂: 4.2; K⁺: 0.6 mmol_c dm⁻³; Ca²⁺: 6 mmol_c dm⁻³; Mg²⁺: 2 mmol_c dm⁻³; H+Al: 38 mmol_c dm⁻³; cation exchange capacity (CIC): 47 mmol_c dm⁻³; and percent base saturation (PSB): 18 %.

Weighing of chemical fertilizers and application in pots

A four-decimal place precision analytical balance was used for weighing the chemicals. Based on the results of the chemical analysis of the soil, calcium carbonate and magnesium carbonate (2.5067 g CaCO₃ and 1.2157 g MgCO₃) were applied in a Ca:Mg ratio of 2:1, in order to increase the percentage of base saturation of the soil to 50 %. Phosphate fertilizer (Agroadubo) was applied in each treatment as indicated: Dose 0:

no phosphorus; Dose 25: 0.6944 mg P dm⁻³; Dose 50: 1.3888 mg P dm⁻³; Dose 75: 2.0832 mg P dm⁻³; Dose 100: 2.7776 mg P dm⁻³; Dose 125: 3.4720 mg P dm⁻³; and Dose 150: 4.1664 mg P dm⁻³. These doses were mixed with carbonates and soil. Subsequently, the soil was transferred to 7 dm³ plastic pots to start the 30-day incubation period.

Ten days after the start of incubation, the following doses of nutrients were applied to all treatments: 15 mg N dm⁻³ (ammonium sulphate); 15 mg K dm⁻³ (potassium chloride); 0.5 mg B dm⁻³ (boric acid); 1.0 mg Zn dm⁻³ (zinc sulphate); 0.5 mg Cu dm⁻³ (copper sulphate); 1.5 mg Mn dm⁻³ (manganese chloride) and 0.02 mg Mo dm⁻³ (ammonium molybdate), through solution and respecting 20 d more to complete the incubation period. After 30 d had elapsed, the soil was carefully removed from the pots individually and dried on plastic sheeting inside the greenhouse. Then, each treatment was homogenized and sampled for chemical analysis of P consisting of ion exchange resin extraction for P and determined by flame spectrophotometry (Raij *et al.*, 2001).

Installation, conducting and termination of the experiment

The plants were established with the roots attached to the substrate of the conical plastic tubes, since this species does not withstand bare root transplanting. Daily irrigations were applied considering the weight of the soil in each pot and considering the daily water loss of the soil contained in the pots due to evapotranspiration. In this way, soil moisture was maintained at 70 % of field capacity.

The experiment was conducted for a period of 90 d. Plant height was recorded at 30, 60, 75 and 90 d after transplanting, using a 20 m tape measure with an accuracy of 8.6 mm at every 2.1 mm. On day 90, the base collar diameter of the main stem of the plants was determined with a digital vernier (0.01 mm). The plants were then cut to 3 cm above the diameter of the root-neck and placed in plastic bags with prior identification, transported to the laboratory and separated into leaves, branches and stems. The roots were extracted from the pots, washed, and sifted to avoid losses.

The total leaf area was determined with the use of an electronic device with Delta-T Devices image analysis system. All samples were washed with water and neutral detergent in a percentage of 0.1 % (1 mL:1 L of water), to avoid errors in the analyses. The samples were then rinsed four times with demineralized water and dried with absorbent paper. The dry biomass weights of the different plant components, as leaves, stems, branches, and roots were then determined. The samples were placed in paper bags and placed in a forced air circulation oven at 70 °C until they reached constant weight. Once the dry biomass weight of the different parts of the plant had been determined, the leaves were ground using a Wiley type stainless steel mill with a 0.841 mm sieve.

To determine the concentration of P in the leaves, a ground composite sample of 0.25 g was obtained from each replicate and placed in test tubes for analysis. A solution was prepared in the ratio of 2:1 (v:v), perchloric acid and nitric acid (nitric-perchloric); then 3 mL were added to each tube containing the sample to be digested. The determination

of P in the plant material was carried out using metavanadate and ammonium molybdate, according to the methodology described by Malavolta *et al.* (1997).

Leaf phosphorus use efficiency (LUE) was calculated by the ratio:

$$\text{EUP (on the leaves)} = \frac{(\text{PS})^2}{\text{CPH}}$$

where: PS = dry biomass weight and CPH = phosphorus content in leaves; CPH was estimated considering leaf dry biomass weights and leaf phosphorus (P) concentrations. The data were subjected to analysis of variance, regression and the mean comparison was done by Tukey test at ($p \leq 0.05$).

RESULTS AND DISCUSSION

Phosphorus content and plant height

When analysing the F values in Table 1, it can be observed that the factor “Seed trees” had highly significant effects ($p \leq 0.01$) on plant height in all measurements made, while the factor “P dose” significantly ($p \leq 0.05$) affected this variable. It was interesting to note that the interaction between factors had no significant effect ($p > 0.05$) on this variable.

The analysis of the factor “seed trees” showed that in the first measurements taken 30 days after the application of the treatments (ddat), the plants from seeds of tree 21

Table 1. Analysis of variance for percentage of phosphorus in the soil after incubation for 30 d and means for plant height as a function of P doses at 30, 60, 75 and 90 d after transplanting, from four seed trees of *Corymbia citriodora*.

Sources of variation	F values			
	30 d	60 d	75 d	90 d
Seed trees	6.35**	12.67**	9.46**	5.00**
P dose	2.36*	2.47*	2.97*	3.49**
Seed trees × P dose	1.19 ^{ns}	1.47 ^{ns}	1.19 ^{ns}	1.14 ^{ns}
CV (%)	15.60	17.15	15.37	14.34

Seed trees	Plant height (cm)			
	30 d	60 d	75 d	90 d
4	13.84 c	39.92 bc	46.91 a	52.51 ab
7	15.70 ab	44.92 ab	49.27 a	54.50 a
21	16.23 a	47.42 a	51.19 a	55.30 a
30	14.38 bc	36.60 c	41.46 b	48.17 b

Means followed by different letters in each column, for each evaluation stage, are statistically different (Tukey; $p \leq 0.05$). Significant at * $p \leq 0.05$. ** $p \leq 0.01$; ns = not significant ($p > 0.05$). CV = coefficients of variation.

showed greater height than the other trees, except for those of Tree 7 (Table 1). The same trend was observed in measurements taken at 60 days. At 75 and 90 ddat, the means of plants from trees 4, 7 and 21 were statistically similar, and the lowest was shown by Tree 30, which at 90 ddat was statistically similar to that shown by Tree 4. Genetic variability among individuals of the same species could be one of the main causes of these differences.

P application differentially affected plant height. Between 30 and 60 d, it was verified that there was an effect of applied P doses on plant height (Table 2).

The unfolding of degrees of freedom for the factor "P dose" in height of *C. citriodora* shows increasing and linear effects with very similar equations, which shows very significant effects with quadratic equations for seed trees 21 at 60 and 75 d of growth, and 30 at 60, 75 and 90 d (Table 2).

Table 2. Regression analysis and means of the percentages of phosphorus in the soil 30 days after incubation in regard to plant height of four seedlings of *Corymbia citriodora* at 30, 60, 75 and 90 days after planting.

Seed trees	Regression	F values			
		30 d	60 d	75 d	90 d
4	Linear	0.40 ^{ns}	1.86 ^{ns}	2.96 ^{ns}	3.29 ^{ns}
	Quadratic	1.51 ^{ns}	1.01 ^{ns}	0.75 ^{ns}	0.43 ^{ns}
	Cubic	0.41 ^{ns}	1.99 ^{ns}	0.27 ^{ns}	0.01 ^{ns}
7	Linear	1.35 ^{ns}	0.89 ^{ns}	2.20 ^{ns}	1.01 ^{ns}
	Quadratic	0.01 ^{ns}	0.70 ^{ns}	0.76 ^{ns}	0.71 ^{ns}
	Cubic	1.32 ^{ns}	1.91 ^{ns}	0.69 ^{ns}	0.35 ^{ns}
21	Linear	2.66 [†]	0.02 ^{ns}	0.01 ^{ns}	0.00 ^{ns}
	Quadratic	0.54 ^{ns}	4.99 [*]	4.02 [*]	3.62 ^{ns}
	Cubic	0.03 ^{ns}	0.44 ^{ns}	1.00 ^{ns}	1.65 ^{ns}
30	Linear	0.45 ^{ns}	0.01 ^{ns}	0.46 ^{ns}	0.92 ^{ns}
	Quadratic	3.43 ^{ns}	14.01 ^{**}	14.87 ^{**}	16.20 ^{**}
	Cubic	0.00 ^{ns}	0.25 ^{ns}	0.16 ^{ns}	0.29 ^{ns}

P dose (mg dm ⁻³)	Plant height (cm)			
	30 d	60 d	75 d	90 d
0	15.40	37.83	41.75	46.30
25	14.34	41.99	46.37	52.64
50	16.10	45.48	50.59	56.34
75	14.23	41.14	46.16	51.06
100	16.05	45.43	50.75	55.53
125	13.85	43.79	48.63	55.38
150	15.30	39.95	46.21	51.09

Means followed by different letters in each column, for each evaluation stage, are statistically different (Tukey; $p \leq 0.05$). Significant at * = $p \leq 0.05$. ** = $p \leq 0.01$; ns = not significant ($p > 0.05$).

In some species of the genus *Eucalyptus* established in clay soils, the application of 60 mg P dm⁻³ is below the critical level for their development and initial growth (Novais *et al.*, 1986).

The positive responses obtained with the application of P demonstrate that this nutrient is indispensable for the initial growth of *C. citriodora*. Most tree species respond to fertilization with increasing doses of macronutrients, which produce favourable percentages in terms of morphological, nutritional evaluations, especially in terms of root expansion (Oliet *et al.*, 2016, Razaq *et al.*, 2017).

Structural carbohydrate allocation is an attribute with strong genetic control; although it may vary according to different tree growth stages, environmental conditions, and planting density (Schumacher *et al.*, 2019). Nutrient uptake capacity and growth response may vary among species, provenances, progenies, and forest clones. Values attributed to genetic variability within the species were observed in the seed trees used in this study.

Diameter of main stem base, leaf area and dry biomass

In this study, significant effects of the factors "Seed trees" and "P dose" were observed for all the variables studied (Table 3), and also the interaction between these two factors studied in regard to dry biomass production of leaves and roots (Table 4).

Table 3. Analysis of variance for plant collar diameter (D), leaf area (AF), leaf dry biomass (BSH), stem and branch dry biomass (BSTR), and root dry biomass (MSR) of plants from four seed trees of *Corymbia citriodora* as a function of phosphorus doses at 90 d after planting.

Sources of variation	F values				
	D	AF	BSH	BSTR	BSR
Seed trees	5.78**	6.38**	14.28**	5.86**	13.03**
P dose	2.97*	4.79**	5.33**	8.70**	2.54*
Seed trees × P dose	1.58 ^{ns}	0.78 ^{ns}	1.86*	1.47 ^{ns}	1.77*
CV (%)	12.43	25.41	22.90	23.58	30.80

Seed trees	Means				
	D (mm)	ÁF (cm ²)	MSH (g)	MSTR (g)	MSR (g)
4	4.94 a	915.50 ab	7.08 bc	5.00 ab	4.65 b
7	4.92 a	998.60 a	8.12 ab	5.31 ab	5.01 ab
21	4.91 a	1076.60 a	8.89 a	5.80 a	5.83 a
30	4.38 b	807.79 b	6.05 c	4.47 b	3.43 c

D: Diameter of the base of the main stem of the plant; AF: leaf area; BSH: dry biomass of leaves; BSTR: dry biomass of stems and branches; BSR: dry biomass of roots. Means followed by different letters, in each column, for each characteristic evaluated, differ statistically (Tukey; $p \leq 0.05$). Significant at * $p \leq 0.05$. ** $p \leq 0.01$; ns = not significant ($p > 0.05$). CV = coefficients of variation.

Table 4. Regression analysis and means of plant collar diameter (D), leaf area (AF), leaf dry biomass (BSH), stem and branch dry biomass (BSTR), and root dry mass (BSR) of plants from four seed trees of *Corymbia citriodora* as a function of phosphorus doses at 90 d after planting.

Seed trees	Regression	F values				
		D	ÁF	MSH	MSTR	MSR
4	Linear	2.32 ^{ns}	7.43 ^{**}	10.95 ^{**}	8.49 ^{**}	3.03 ^{ns}
	Quadratic	0.06 ^{ns}	0.25 ^{ns}	0.32 ^{ns}	0.00 ^{ns}	0.02 ^{ns}
	Cubic	3.88 ^{ns}	0.42 ^{ns}	0.07 ^{ns}	0.37 ^{ns}	1.81 ^{ns}
7	Linear	1.95 ^{ns}	0.92 ^{ns}	1.32 ^{ns}	6.2 [*]	0.56 ^{ns}
	Quadratic	3.07 ^{ns}	0.24 ^{ns}	4.08 [*]	6.76 [*]	3.76 ^{ns}
	Cubic	3.22 ^{ns}	0.01 ^{ns}	2.74 ^{ns}	1.07 ^{ns}	5.29 [*]
21	Linear	0.04 ^{ns}	6.09 [*]	1.58 ^{ns}	2.70 ^{ns}	0.25 ^{ns}
	Quadratic	7.58 ^{**}	8.49 ^{**}	5.25 [*]	10.44 ^{**}	4.60 [*]
	Cubic	1.23 ^{ns}	2.07 ^{ns}	3.87 ^{ns}	2.04 ^{ns}	0.12 ^{ns}
30	Linear	0.07 ^{ns}	4.91 [*]	0.05 ^{ns}	1.14 ^{ns}	0.92 ^{ns}
	Quadratic	4.75 [*]	4.64 [*]	15.77 ^{**}	20.93 ^{**}	6.83 [*]
	Cubic	0.06 ^{ns}	0.08 ^{ns}	0.00 ^{ns}	0.06 ^{ns}	0.59 ^{ns}

P dose (mg dm ⁻³)	Means				
	D (mm)	AF (cm ²)	BSH (g leaf ⁻¹)	BSTR (g leaf ⁻¹)	BSR (g leaf ⁻¹)
0	4.36	698.69	5.64	3.44	3.98
25	4.73	836.88	7.08	4.73	4.47
50	5.11	1033.62	8.79	6.01	5.64
75	4.75	990.43	7.58	5.24	4.87
100	4.97	1038.43	8.27	5.83	5.04
125	4.99	1047.38	7.81	5.82	5.00
150	4.61	1001.93	7.60	4.95	4.14

D: Diameter of the base of the main stem of the plant; AF: leaf area; BSH: dry biomass of leaves; BSTR: dry biomass of stems and branches; BSR: dry biomass of roots. Means followed by different letters in each column, for each evaluation stage, are statistically different (Tukey; $p \leq 0.05$). Significant at * = $p \leq 0.05$. ** = $p \leq 0.01$; ns = not significant ($p > 0.05$).

Coefficient of variation values were considered medium for main stem base diameter, high for leaf area, high for dry biomass of leaves, stems, and branches, and very high for dry biomass of roots (Table 3). These higher coefficient of variation values for root biomass weight may be associated with genetic variability among seed trees.

Trees 4, 7 and 21 produced plants with stem diameter similar to each other, and superior to those of tree 30, whose plants had the smallest diameter. Similarly, trees 4, 7 and 21 produced plants with similar leaf area to each other, although the leaf area of plants from Tree 4 was statistically similar to those from Tree 30. The highest means of leaf dry biomass weight (BSH) were observed in plants produced by trees 21 and 7, and the lowest mean in plants from trees 4 and 30; trees 4 and 7 showed plants with statistically similar means for this variable. For dry biomass weight of stems and

branches (BSTR), the plants of trees 4, 7 and 21 showed similar means to each other, and those of tree 30 were the lowest, also statistically similar to those shown by the plants of trees 4 and 7. The weight of root dry biomass (BSR) was higher in plants from trees 7 and 21, and lower in plants from tree 30; the means of plants from trees 4 and 7 were statistically similar (Table 3).

An exhaustive search in the specialized literature did not find any research on the effect of phosphate fertilization on this type of forest trees. In fact, the greatest difficulty for the design of new experiments is the lack of studies that provide previous and detailed information on the selection of matrix trees (Barreiros *et al.*, 2002). In *Pinus greggii* var. *greggii*, differential nitrogen and potassium supply was found to significantly affect leaf phosphorus concentration (Vázquez-Cisneros *et al.*, 2018). Phosphorus is stored in the leaves of the plant, differing according to the seed tree, probably due to physiological and genetic characteristics of the plant.

The “luxury consumption” of a fertilizer (increased availability of a nutrient that does not generate an increase in growth rate) may not be detrimental to the plant, although it represents a waste of this input and an unnecessary expense (Massone *et al.*, 2018). Therefore, this research determined the dose that is necessary for the growth of *C. citriodora* to avoid waste or phytotoxicity.

There was no evidence that the roots of *C. citriodora* plants had grown more than the aerial part, based on biomass weight data. In plants from seed of Tree 21, a higher phosphorus requirement was observed (about 100 mg P dm⁻³) to obtain maximum leaf area, maximum dry biomass production of leaves, stems, and branches, compared to dry biomass production of roots (about 65 mg P dm⁻³). This indicates that this genetic material is efficient in phosphorus uptake and biomass production. The analysis of seed from trees 21 and 30 showed positive and significant linear regressions, while the quadratic regression of seed from trees 4 and 7 also shows significant effects for some organs, but not for the root (Table 4). In *Euterpe edulis*, it was observed that the leaf was the second organ to accumulate nutrients by supplying 26.2 % phosphorus in 2, 4, 6, and 8-year-old trees (Schumacher *et al.*, 2019). This statement is similar to the results obtained in this research, only varying the age of the plants, which was 6 months.

The diameter of the base of the main plant collar showed a quadratic effect for seed trees 21 and 30 (Table 4). This growth variable, besides being influenced by fertilization, is widely used as an indicator of forest tree quality.

Phosphorus contributes to optimize plant morphological measurements, increase internal nutritional percentages, and stimulate root system development (Razaq *et al.*, 2017). In this study the above ground part developed more than the roots, due to phosphate fertilization (Table 3).

In *Psidium guajava*, phosphorus and nitrogen deficiency decreased leaf area and leaf number (Dussan *et al.*, 2016). In pot-grown clones of *Eucalyptus dunnii*, and *Eucalyptus benthamii*, the addition of phosphorus to the soil increased leaf dry biomass production and interfered with parameters related to nutritional efficiency (Stahl *et al.*, 2013).

Phosphorus concentration, content and use efficiency in leaves

There was variation among seed trees and among phosphorus treatments for phosphorus content and use efficiency in leaves of *C. citriodora*.

Tree 30 was the least efficient in phosphorus use compared to the other seed trees, while tree 21 presented the highest mean for this variable, with 29.38 mg P dm⁻³. Tree 30 presented the highest mean for foliar phosphorus concentration with the dose of 57.25 mg P dm⁻³, and for phosphorus use efficiency and of seed Tree 30 the efficiency was estimated at 3623.46, with the dose of 61.68 mg P dm⁻³ (Table 5).

Despite the existence of a significant effect for the interaction between factors for accumulated phosphorus content in leaves per plant, the analysis of degrees of freedom was done to study the effects of phosphorus doses with phosphorus concentration, content in milligrams and efficiency of phosphorus absorption by leaves for the seed trees studied. According to the regression analysis performed for all the variables studied, such as P concentration and content, as well as phosphorus use efficiency, it was linear, except for trees 4 and 30; however, these obtained positive quadratic regression. The Tree 21 plants showed significant effects in all the characteristics evaluated (Table 6).

Variations among seed trees in relation to the amounts and efficiencies of phosphorus use are related to genetic variability among seed trees and the results show that seed tree 21 was the most efficient in phosphorus use for leaf biomass production. In the

Table 5. Analysis of variance of phosphorus concentration, phosphorus content and phosphorus use efficiency in leaves of plants from four *Corymbia citriodora* seed trees as a function of phosphorus doses, 90 d after transplanting.

Sources of variation	F values		
	Concentration of P	Content of P	Phosphorus use efficiency
Seed trees	21.61**	2.80*	23.02**
P dose	35.23**	22.02**	5.32**
Seed trees × P dose	1.06 ^{ns}	2.78**	1.56 ^{ns}
CV (%)	12.83	22.66	28.74

Seed trees	Means		
	Concentration of P (g kg ⁻¹ of dry matter)	Content of P (mg leaf ⁻¹)	Phosphorus use efficiency
4	1.58b	11.45 ab	4582.78 b
7	1.41c	11.40 ab	6064.46 a
21	1.39c	12.52 a	6588.60 a
30	1.78a	10.51 b	3623.46b

Means followed by different letters, in each column, for each characteristic evaluated, are statistically different (Tukey; $p \leq 0.05$). Significant at * $p \leq 0.05$. ** $p \leq 0.01$; ns = not significant ($p > 0.05$). CV = coefficients of variation.

Table 6. Regression analysis for phosphorus concentration, phosphorus content and phosphorus use efficiency in leaves of plants from four *Corymbia citriodora* seedlings as a function of phosphorus doses, 90 d after transplanting.

Seed trees	Regression	F values		
		Concentration of P	Content of P	Phosphorus use efficiency
4	Linear	44.21**	46.69**	0.06 ^{ns}
	Quadratic	3.18 ^{ns}	0.02 ^{ns}	1.46 ^{ns}
	Cubic	0.01 ^{ns}	0.29 ^{ns}	0.28 ^{ns}
7	Linear	38.91**	24.02**	7.96**
	Quadratic	0.12 ^{ns}	3.93 ^{ns}	1.34 ^{ns}
	Cubic	2.06 ^{ns}	0.64 ^{ns}	3.59 ^{ns}
21	Linear	57.34**	35.37**	10.74**
	Quadratic	0.15 ^{ns}	1.89 ^{ns}	4.10*
	Cubic	0.45 ^{ns}	0.57 ^{ns}	5.64*
30	Linear	52.11**	7.88**	2.10 ^{ns}
	Quadratic	4.44*	24.83**	5.56**
	Cubic	0.54 ^{ns}	0.35 ^{ns}	0.01 ^{ns}

P dose (mg dm ⁻³)	Means		
	Concentration of P (g kg ⁻¹ of dry matter)	Content of P (mg leaf ⁻¹)	Phosphorus use efficiency
0	1.09	5.93	5469.00
25	1.34	9.33	5524.43
50	1.38	11.90	6676.62
75	1.56	11.49	5146.81
100	1.64	13.04	5312.87
125	1.95	15.27	4019.12
150	1.80	13.34	4354.93

Means followed by different letters in each column, for each evaluation stage, are statistically different (Tukey; $p \leq 0.05$). Significant at * = $p \leq 0.05$. ** = $p \leq 0.01$; ns = not significant ($p > 0.05$).

initial stage of plant development, there is the probability of high phosphorus demand, as found by (Leite *et al.*, 2006).

The concentration and amount of phosphorus in the leaves increased with increasing phosphorus doses. For seed tree 21, there was a third-degree effect of phosphorus doses for leaf phosphorus utilization efficiency. However, with doses higher than 30 mg P dm⁻³ there was a reduction in the efficiency of utilization of this chemical element. When a plant is in a situation of low phosphorus availability, it tends to use it more efficiently, contrary to what is observed when it is subjected to conditions of higher fertility (Stahl *et al.*, 2013).

In this research Tree 21 was the most efficient in phosphorus use (Table 5). The potential of the same genotype to produce different phenotypes in response to

different environmental conditions is one of the means by which plants can adjust their morphology and physiology to cope with environmental heterogeneity (Gianoli, 2004).

High doses of phosphorus (300 mg L^{-1}) during the maturity stage of *Aextoxicon punctatum* plant tissues produce the highest foliar nutritional concentrations of nitrogen and the lowest of calcium (González *et al.*, 2020). In this study, it was observed that the treatments with the optimal dose were more efficient in the transport of low-available phosphate ion into the plant.

The small variation in phosphorus foliar concentration in relation to the applied doses demonstrates the occurrence of a dilution effect, provided by the higher plant production with phosphorus application, with greater production of leaf biomass than stems and branches (Table 5). In a study with four *Pinus* families and phosphorus fertilization, P increased plant growth in all four families, and phosphorus fertilization changed some hydraulic parameters, but these changes did not affect growth (Faustino *et al.*, 2013). Furthermore, it was observed that as plants get older, the increased capacity of the root system to explore larger amounts of soil and consequently larger amounts of phosphorus for growth.

Seed trees that produce greater volume of wood, show greater growth in height and export different amounts of phosphorus from the soil to the above ground part of the plant, being the most suitable due to the limitation of this nutrient in soils destined for plantations. The use of seed trees explores the possibility of reducing the manifestation of this genetic heterogeneity between seed trees in a forest and plantations. In a study with *Parapiptadenia rigida*, higher growth was observed in young plants at a dose of 450 mg P kg^{-1} (Schumacher *et al.*, 2004). These studies can be used to recommend an accurate dose at a young stage in the growth of trees, in order to reduce costs and misuse of phosphorus.

CONCLUSIONS

Phosphate fertilization is necessary for the growth and development of *Corymbia citriodora* plants. The four *C. citriodora* seed trees from different geographical areas in São Paulo, Brazil, responded differently to the application of different increasing doses of phosphorus. Consequently, according to the characteristic evaluated and the seed tree, it was found that the adequate amount of phosphorus-based chemical fertilizer is between 30 and 100 mg dm^{-3} for the initial production of high-quality plants because it is efficient and sustainable.

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REFERENCES

- Barreiros RM, Garcia JN, Caixeta Filho JV, Sansigolo CA. 2002. Optimization model for selection of *Eucalyptus grandis* matrix trees. *Forest Sciences* 30: 25–39.
- Ciavatta SF, da Silva MR, Simões D. 2014. Fertirrigation in production of seedlings of *Eucalyptus grandis* during winter and summer. *Cerne* 20 (2): 217–222. <https://doi.org/10.1590/01047760.201420021374>
- Dussan SL, Villegas DA, Miranda D. 2016. Efecto de la deficiencia de N, P, K, Mg, Ca y B sobre la acumulación y distribución de la masa seca en plantas de guayaba (*Psidium guajava* L.) var. ICA Palmira II en fase de vivero. *Revista Colombiana de Ciencias Hortícolas* 10 (1): 40–52. <https://doi.org/10.17584/rcch.2016v10i1.4277>
- FAO (Organización de las Naciones Unidas para la Alimentación y la Agricultura). 2015. World fertilizer trends and outlook to 2018. Roma, Italia. http://agricultura.gencat.cat/web/.content/de_departament/de02_estadistiques_observatoris/27_butlletins/02_butlletins_nd/documents_nd/fitxers_estadistics_nd/2015/0153_2015_SProductius_Fertilitzacio_Fertilitzants-Mon.pdf (Retrieved: January 2022).
- Faustino LI, Bulfe NML, Pinazo MA, Monteoliva SE, Graciano C. 2013. Dry weight partitioning and hydraulic traits in young *Pinus taeda* trees fertilized with nitrogen and phosphorus in a subtropical area. *Tree Physiology* 33 (3): 241–251. <https://doi.org/10.1093/treephys/tps129>
- Filho FB, Ferreira ME, Vieira RD, Pessôa da Cruz MC, Pessôa da Cruz C MA, de Barros S T, C Lopes R JG. 2013. Adubação com fósforo e potássio para produção e qualidade de sementes de soja. *Pesquisa Agropecuária Brasileira* 48 (7): 783–790. <https://doi.org/10.1590/S0100-204X2013000700011>
- Gianoli E. 2004. Plasticity of traits and correlations in two populations of *Convolvulus arvensis* (Convolvulaceae) differing in environmental heterogeneity. *International Journal of Plant Sciences* 165 (5): 825–832. <https://doi.org/10.1086/422050>
- González M, Ríos D, Peña-Rojas K, García E, Acevedo M, Cartes E, Sánchez-Olate M. 2020. Efecto de la concentración de fósforo y calcio sobre atributos morfo-fisiológicos y potencial de crecimiento radical en plantas de *Aextoxicon punctatum* producidas a raíz cubierta en la etapa de endurecimiento. *Bosque (Valdivia)* 41 (2): 137–146. <https://doi.org/10.4067/S0717-92002020000200137>
- Leite PB, Alvarez VVH, Barros NF, Neves JCL, Guarçoni MA. 2006. Níveis críticos de fósforo, para milho, em casa de vegetação, de acordo com a sua localização no solo. *Revista Brasileira de Ciência do Solo* 30 (3): 497–508.
- Lima AMN, Neves JCL, Silva IR, Leite FP. 2005. Cinética de absorção e eficiência nutricional de K⁺, Ca²⁺ e Mg²⁺ em plantas jovens de quatro clones de eucalipto. *Revista Brasileira de Ciência do Solo* 29 (6): 903–909. <https://doi.org/10.1590/S0100-06832005000600008>
- Malavolta E, Vitti EC, Oliveira SA. 1997. Avaliação do estado nutricional das plantas: princípios e aplicações (2nd Edition); Associação Brasileira para Pesquisa da Potassa e do Fosfato: São Paulo, Brasil.
- Massone DS, Bartoli CG, Pastorino MJ. 2018. Efecto de la fertilización con distintas concentraciones de nitrógeno y potasio en el crecimiento de plantines de ciprés de la cordillera (*Austrocedrus chilensis*) en vivero. *Bosque (Valdivia)* 39 (3): 375–384. <https://doi.org/10.4067/S0717-92002018000300375>
- Morais E, Zanatto ACS, Freitas MLM, de Moraes MLT, Sebbenn AM. 2010. Genetic variation, genotype × soil interaction and genetic gains in a *Corymbia citriodora* Hook progeny test in Luiz Antonio, São Paulo, Brazil. *Forest Sciences* 38 (85): 11–18.
- Novais, RF, Barros NF, Neves, JCL. 1986. Interpretação de análise química do solo para o crescimento e desenvolvimento de *Eucalyptus* spp. Níveis críticos de implantação e de manutenção. *Revista Árvore* 10 (1): 105–111.
- Oliet JA, Planelles R, Artero F, Domingo-Santos JM. 2016. Establishing *Acacia salicina* under dry Mediterranean conditions: The effects of nursery fertilization and tree shelters on a mid-term experiment with saline irrigation. *Ciencia e Investigación Agraria* 43 (1): 69–84. <https://doi.org/10.4067/S0718-16202016000100007>
- Oliveira JG, Silva VSG, Costa JPV. 2017. Comportamento de soja submetida a materiais fertilizantes e inoculação com bradyrhizobium. *Revista da Universidade Vale do Rio Verde, Três Corações*. 15 (1): 66–72. <https://doi.org/10.5892/ruvrd.v15i1.3016>
- Raij BV, de Andrade JC, Cantarella H, Quaggio JA (eds.) 2001. Análises químicas para avaliação da fertilidade de solos tropicais. Campinas, Instituto Agrônomo: São Paulo, Brasil.

- Razaq M, Zhang P, Shen H, Salahuddin. 2017. Influence of nitrogen and phosphorous on the growth and root morphology of *Acer mono*. PLoS ONE 12: e0171321. <https://doi.org/10.1371/journal.pone.0171321>
- Schumacher MV, Witschoreck R, Neves CF, Lopes VG. 2019. Manejo da biomassa e sustentabilidade nutricional em povoamentos de *Eucalyptus* spp. em pequenas propriedades rurais. Ciência Florestal 29 (1): 144–156. <https://doi.org/10.5902/198050985135>
- Schumacher MV, Ceconi DE, Santana CA. 2004. Influence of different phosphorus doses on the growth of angico-vermelho seedlings (*Parapiptadenia rigida* (Benth) Brenan). Revista Árvore 28 (1): 149–155. <https://doi.org/10.1590/S0100-67622004000100019>
- Stahl J, Ernani PR, Gatiboni LC, Chaves DM, Neves CU. 2013. Produção de massa seca e eficiência nutricional de clones de *Eucalyptus dunnii* e *Eucalyptus benthamii* em função da adição de doses de fósforo ao solo. Ciência Florestal 23 (2): 287–295. <https://doi.org/10.5902/198050989275>
- Vázquez-Cisneros I, Prieto-Ruiz JA, López-López MA, Wehenkel C, Domínguez-Calleros PA, FE Muñoz-Sáez. 2018. Growth and survival of a plantation of *Pinus greggii* Engelm. ex Parl. var. *greggii* under different fertilization treatments. Revista Chapingo Serie Ciencias Forestales y del Ambiente 24 (2): 251–264. <https://doi.org/10.5154/r.rchscfa.2017.05.036>