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Quality of eucalyptus seedlings of different clones produced at different times

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ABSTRACT

Fast-growing economic forests are important, especially for the cultivation of eucalyptus, as they represent multiple uses associated with the preservation of native forests, generation of jobs, and income. In recent years, there has been significant growth in the number of nurseries that produce clonal eucalyptus seedlings throughout Brazil, and in Espírito Santo soil, most are concentrated in Sooretama, producing seedlings throughout Brazil and for export. However, information on the quality of these seedlings when produced throughout the different seasons is not available. This information can be important, from a practical point of view, for planting and management, and from an experimental point of view, to direct research involving newly produced seedlings. Thus, this work aims to qualify, through the characteristic of DQI, clonal eucalyptus seedlings of different clones and in different seasons, to study the presence of interaction between the clones, and the environments formed by the different seasons of the year. Statistical analyzes were performed from 6,400 eucalyptus seedlings of four clones produced at different times in Sooretama and Jaguaré in the state of Espirito Santo, applying joint evaluation methods to study the interaction between genotypes by environments and verification of adaptability and stability. In the analysis by Annicchiarico and Lin & Binns, the G3 genotype showed general adaptability and also showed adaptability in unfavorable environments, and the G2 and G4 genotypes showed adaptability to favorable environments. Despite the specificities of adaptability, all four genotypes can be indicated for planting at any time of the year, as they have an DQI greater than 0.2.

Key words: *Eucalyptus urograndis*. *Eucalyptus urophylla*. Clonal seedlings. Statistical analysis. Interaction between genotypes and environments. Eucalyptus cultivation. Dickson's Quality Index. Adaptability and Stability.

Adherence to the scope of BJEDIS: This is a research with a significant amount of eucalyptus seedlings and that, to measure the quality, makes use of several biometric resources.

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1. INTRODUCTION

With a leading position in the development of a low carbon economy, the forest-based sector stands out for its high productivity, embedded technology, best forest management practices, social responsibility, and modern production facilities (1). Of great economic importance, representing 1.2% of the National GDP and total gross revenue of R$ 97.4 billion, this is an industry with an eye on the future, which invests in research to develop products that are aligned with the bioeconomy (2). This vision has allowed growth in the sector, generating opportunities for millions of Brazilians.

In 2020, the Eucalyptus plantation area in Brazil totaled 7.47 million hectares, representing 78% of the total planted area in Brazil of trees, concentrated mainly in the States of Minas Gerais, São Paulo, and Mato Grosso do Sul, with the State of Espírito Santo, the planted area was 230,918 hectares (2). Planning carried out by the Espírito Santo Department of Agriculture for the period between 2007 and 2025 indicates that the aim is to increase the planting area of fast-growing economic forests to 512,700 hectares by 2025, with their cultivation being encouraged, mainly in the recovery and conservation of marginal areas, counting on the expansion of research actions, both for the segment of large producers and for family-based farmers (3).

Despite advances in research and cultural techniques with eucalyptus in Brazil, the production of seedlings remains one of the main problems in the implementation of new crops (4-5-6-7). These problems are aggravated by the interaction between the different genotypes and the environmental conditions throughout the different seasons of the year, especially temperature, light, photoperiod, and relative humidity (7).

Some studies have been carried out to evaluate the quality of seedlings, in tubes, most of them with *Eucalyptus grandis* and by seminiferous propagation (8-9-10). However, recently, there has been a preference for clonal plantations, due to their greater volumetric increment, uniform plant growth, disease control, and raw material homogeneity (11). It appears that few works are related to the production of clonal seedlings of the most recent hybrids of *Eucalyptus urograndis* (*E. grandis* x *E. urophylla*) (12). Another observation that is made is that the experimental evaluations are based on a single time (station) of data collection, not knowing the behavior along the seasons.

The Dickson Quality Index is one of the quality indicators of seedlings since it takes into account several important morphological indicators at the same time so that the use of these parameters in isolation can increase the risk of the wrong choice of taller seedlings. to the detriment of the lowest (13). Taller seedlings are often not the best in terms of field survival. Hunt (14) proposed a minimum value for the DQI as a good indicator of seedling quality, being 0.20. Concerning the quality of eucalyptus seedlings, its use has been vast, including studies with seedlings of seminiferous propagation (3-15) and with seedlings of clonal propagation (16-17).

It is of fundamental importance to know if there are statistical differences between the clones and between them and the production times of eucalyptus seedlings, which is called genotype by environment interaction (18). The possible differences detected may be an important indication of the need for different management and cultural treatments when planting different clones at different times of the year.

Thus, the objective of this work was to evaluate seedlings of the main eucalyptus clones produced in tubes, in the four seasons of the year and two years, in the north of Espírito Santo, aiming to evaluate the presence of genotypes x environments interaction, based on the analysis of stability and adaptability for the DQI characteristic.

2. MATERIALS AND METHOD

The seedlings evaluated statistically were obtained from commercial nurseries in Sooretama and Jaguaré, with clonal eucalyptus seedlings, with 90 days and between 20 and 30 cm in height, kept in 10 x 14 trays containing 50 seedlings. The seedlings are from clones 224 (*Eucalyptus urophylla*), BA7346, CO1407, and TP361 (*Eucalyptus urograndis*), as shown in Table 1. The evaluations were carried out at the end of each season, expressing the result of the environmental action suffered during the three months of training. They were evaluated: in spring, summer, autumn, and winter, according to Table 2.

Table **1**. List of eucalyptus genotypes, their respective species and acronyms.

Genotypes	
AEC 244 -*Eucalyptus urophylla*	G1
BA 7346 -*Eucalyptus urograndis*	G2
CO 1407 -*Eucalyptus urograndis*	G3
TP361 -*Eucalyptus urograndis*	G4

Table **2**. Assessed environments, assessment times and development season.

Environments	Evaluation times	Seedling development season
1	December 2015	Spring
2	March 2016	Summer
3	June 2016	Autumn
4	September 2016	Winter
5	December 2016	Spring
6	March 2017	Summer
7	June 2017	Autumn
8	September 2017	Winter

In each clone and each station, 200 seedlings were evaluated, identified from 1 to 200. Thus, during the research, between 2015 and 2017, the following characteristics were evaluated, to later calculate the Dickson Quality Index (DQI) (19), in 6400 seedlings: height (H, in cm), measured with a ruler from the base on the substrate until the insertion of the last leaf; stem diameter (CD, in mm), measured with a digital caliper at the base of the collar; shoot dry mass (SDM, in g); root dry mass (RDM, in g) and total dry mass (TDM, in g). The characters SDM and RDM were determined on an analytical balance, after drying in an oven at 60 ºC until constant weight. The Dickson Quality Index was determined, through expression 1 (19).

$$\mathrm{DQI} = \frac{\mathrm{TDM(g)}}{\left(\frac{\mathrm{H(cm)}}{\mathrm{CD(mm)}}\right)+\left(\frac{\mathrm{SDM(g)}}{\mathrm{RDM(g)}}\right)} \qquad \text{Expression 1}$$

From the data of the characteristics previously evaluated in each of the 200 seedlings of each of the four clones in the four seasons in the two years, different statistics were performed, the data were submitted to simple factorial ANOVA, with 5 replications, in the fixed genotype model and random environment. Assumptions of normality, homogeneity of variances and sphericity were satisfied To verify if there is an interaction between genotypes and environments, a joint analysis of the variance of experiments was performed (20-21). Once the interaction was detected, it was necessary to take measures to alleviate the problems that this interaction can cause (18-21). With the existence of the interaction between genotypes and environments, studies of adaptability and stability were carried out using the Annicchiarico method (22) modified by Schmildt & Cruz (23). For comparison purposes, the methodology proposed by Lin & Binns (24) and the modifications proposed by Carneiro (25) were used. For all statistical analyses, the Genes program was used (26).

By the method of Annicchiarico (22), stability is measured by the superiority of the genotype to the mean of each environment. The method is based on the estimation of a confidence index for a given genotype to show relatively superior behavior (27). By this method, the confidence index is obtained by the following expression 2:

$$I_i = \overline{Y}_{i.} - Z_{(1-\alpha)}(\hat{\sigma}_{i.}) \qquad \text{Expression 2}$$

On what:

I_i: confidence index;

$\overline{Y}_{i.}$: average of genotype i in percentage in relation to the average of all environments;

$Z_{(1-\alpha)}$: value of the standardized normal distribution in which the cumulative distribution function meets the value $(1-\alpha)$, with a significance level α pre-set by the author at 0.25.

$\hat{\sigma}_{i.}$: standard deviation of the mean of cultivar i in percentage in relation to all environments.

Annicchiarico's method (22) as reported in expression 2 assesses broad or general adaptability. When the equation is used in relation to favorable or unfavorable environments, the classification of these environments is made by the environmental index (Ij) according to Schmildt & Cruz (23) and shown in expression 3 and 4:

$$I_{if} = \overline{Y}_{i.} - Z_{(1-\alpha)}(\hat{\sigma}_{if}) \qquad \text{Expression 3}$$

On what:

$\hat{\sigma}_{if}$: standard deviation of the mean of cultivar i in percentage in relation to favorable environments.

$$I_{id} = \overline{Y}_{i.} - Z_{(1-\alpha)}(\hat{\sigma}_{id}) \qquad \text{Expression 4}$$

On what:

$\hat{\sigma}_{id}$: standard deviation of the mean of cultivar i in percentage in relation to unfavorable environments

The method proposed by Lin & Binns (24) allows the measurement of how close the cultivar is to the ideal performance, referred to as a genotype with the best performance in all environments studied (28). The genotypes considered stable are those with the lowestPi, obtained according to expression 5:

$$P_i = \frac{\sum_{i=1}^{n}\left(M_{ij}-M_j\right)^2}{2n} \qquad \text{Expression 5}$$

On what:

P_i: stability index of genotype i;

M_{ij}: mean of genotype i in environment j;

M_j: mean of the genotype with maximum response among all in environment j;

n: number of environments.

The Lin & Binns method (24) as reported in expression 5 assesses broad or general adaptability. When the equation is used for favorable or unfavorable environments, the classification of these environments is made according to Carneiro (25), with the decomposition of the Pi estimator, in the parts due to favorable environments (Pif), expression 6, and unfavorable (Pid), expression 7. The classification of these environments was made based on the environmental indices, defined as the difference between the average of the evaluated cultivars in each environment and the general average of the experiments.

For favorable environments, with indices greater than or equal to zero, Pif was estimated as follows:

$$P_{if} = \frac{\sum_{i=1}^{n}(M_{ij}-M_j)^2}{2f}$$ Expression 6

On what:

f: number of favorable environments;

Y_{ij} e Mj: as defined above.

For unfavorable environments:

$$P_{id} = \frac{\sum_{i=1}^{n}(M_{ij}-M_j)^2}{2d}$$ Expressão 7

On what:

d: number of unfavorable environments.

3. RESULTS AND DISCUSSION

As for adaptability and stability, according to the confidence index proposed by Annicchiarico (22) for environments in general, for the DQI values of the 4 genotypes, G3 presented a performance 10% higher than the environmental average, as observed in Table 3, showing greater stability and general adaptability. Genotypes with above-average behavior present a lower risk of adoption by farmers, as their general adaptability characteristic gives them a greater probability of presenting higher DQI averages in each environment (27). Genotypes G3 and G4 showed stability in unfavorable environments, showing them, with 75% confidence, 29.15%, and 5.08% higher than the average in unfavorable environments, respectively. In favorable environments, the genotypes G2 and G4 presented performances of 6 and 3%, respectively, superior to the average of the favorable environments.

Schmildt & Cruz (23) evaluated different stability methods and concluded that the Annicchiarico method was more reliable than Eberhart and Russell because of the ease of interpretation, the precision in the indication of cultivars for each type of environment, and the fact that the method has only one interpretation parameter while the Eberhart & Russell method takes four parameters.

Environments 2 (seedlings formed in the summer of 2016), 5 (seedlings formed in the spring of 2016), and 6 (seedlings formed in the summer of 2017) were considered unfavorable (Table 4), while the others were considered favorable. This result can be explained by variations in temperature, photoperiod, light, and relative humidity throughout the different seasons. By the Lin & Binns method, the same results are obtained regarding the classification of environments into favorable or unfavorable by the two methodologies used (Table 4).

As the values obtained for adaptability and stability by this method approached 100, the method proposed by Lin & Binns was performed for comparison.

Table **3**. Estimates of the average DQI, of the Confidence Index, according to Annicchiarico (22), with decomposition for favorable and unfavorable environments, according to Schmildt & Cruz (23) in the evaluation of eucalyptus clones.

Genotypes	Average		Confidence index (I_i) according to environments					
			General		Favorable		Unfavorable	
		1/		2/		2/		2/
G1	0.2401	(4)	88.46	(4)	91.67	(4)	83.11	(3)
G2	0.2756	(3)	97.46	(3)	106.35	(1)	82.65	(4)
G3	0.3008	(1)	110.01	(1)	98.52	(3)	129.15	(1)
G4	0.2853	(2)	104.07	(2)	103.46	(2)	105.08	(2)
Average	0.2755		-		-		-	

1/Classification of cultivars by average production.
2/Classification of cultivars, by the values of I_i.

Table **4**. Classification of Environments by the environmental index, used to classify environments into favorable and unfavorable for use in the methods of Annicchiarico (22) modified by Schmildt & Cruz, 23) and Lin & Binns (24) modified by Carneiro (25).

Environment	Class	
	Annicchiarico	Lin & Binns
1	Favorable	Favorable
2	Unfavorable	Unfavorable
3	Favorable	Favorable
4	Favorable	Favorable
5	Unfavorable	Unfavorable
6	Unfavorable	Unfavorable
7	Favorable	Favorable
8	Favorable	Favorable

By the method of Lin & Binns (24) with its modifications proposed by Carneiro (25), it was found that two clones were the most stable for the DQI trait in environments of general adaptability, the genotypes G2 and G3, presented the same value, stood out for having the lowest overall Pi value, being 0.0151, as shown in Table 5. The G4 showed specific adaptability to favorable environments since it was stable in favorable environments. Therefore, because they present these characteristics, they are indicated for cultivation in favorable environments or for farmers with a high level of investment. Stability over the years and environments are preferred, as they tend to present predictable behaviors, which facilitates their indication (29). In unfavorable environments the G3 stood out, indicating that even under adverse conditions this clone presented good quality. Clones with this behavior can be recommended for low-investment farmers or poor and disadvantaged soil locations. G1 was the least stable in all environments, both favorable and unfavorable, which can be explained by the fact that this genotype is the only one that is not a hybrid, but even so, it is still considered to be of good quality because it has an DQI > 0.2 as proposed by Hunt (14).

Table **5**. Estimates of the averages of DQI, of the general P_i, of the favorable and of the unfavorable P_i, by the method of Lin & Binns (24) with decomposition of the parameter P_i according to Carneiro (25), for the eucalyptus genotypes.

Genotypes	Average		General P_i		Favorable P_i		Unfavorable P_i	
		1/		2/		2/		2/
G1	0.2401	(4)	0.0210	(4)	0.0271	(4)	0.0108	(3)
G2	0.2756	(3)	0.0151	(1)	0.0168	(1)	0.0122	(4)
G3	0.3008	(1)	0.0151	(2)	0.0237	(3)	0.0008	(1)

G4	0.2853	(2)	0.0172	(3)	0.0224	(2)	0.0085	(2)
Average	0.2754		-		-		-	

[1/] Classification of cultivars by average production.
[2/] Classification of cultivars, by P_i values.

In this work, the hybrids obtained better results, and according to Paludzyszyn Filho et al. (30), the *E. grandis*, along with the *E. urophylla*, form one of the best hybrid combinations and, under proper management of population density, ensure good productivity of raw material for various uses.

4. CONCLUSION

All genotypes presented DQI value within the standard that demonstrates good quality. The methods by Annicchiarico and Lin & Binns showed equivalent results in indicating the quality of eucalyptus clones. The hybrids (*E. urograndis*) obtained better results than the clone of *E. urophyla*, however, all genotypes showed good quality. Genotypes G2 and G4 are recommended for favorable environments, or high-investment producers, G3 showed general adaptability but also showed the best result in unfavorable environments.

CONFLICT OF INTEREST

There is no conflict of interest.

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Sample CRediT author statement

Ana Valeria Lima Santos: Conceptualization, Methodology, Data analysis, and Writing-Original draft preparation**.** **Rubens Neres Araújo**:.. **Nathan Ribeiro de Castro**:... **Karina Tiemi Hassuda dos Santos**:…**Gleyce Pereira Santos**:... **Vinicius de Souza Oliveira** …**Diana Gonçalves Costa**: …**Jeniffer Ribeiro de Oliveira**:… **Omar Schmildt**: … **Edilson Romais Schmildt:**…

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