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MORPHOPHYSIOLOGICAL QUALITY OF *Ilex paraguariensis* SEEDLINGS PRODUCED IN DIFFERENT SEEDLING PRODUCTION UNITS

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Resumo

Qualidade morfofisiológica de mudas de Ilex paraguariensis produzidas em diferentes unidades de produção de mudas. A erva-mate (*Ilex paraguariensis* St. Hill.) é uma espécie de grande potencial nutracêutico, farmacêutico e alimentício e, por esta razão, suas áreas de cultivo têm sido ampliadas, o que, por sua vez, aumenta a demanda por mudas de qualidade. Nesse sentido, este trabalho teve por objetivo comparar o crescimento e desenvolvimento de mudas de *Ilex paraguariensis* produzidas em três diferentes unidades de produção de mudas (recipiente biodegradável de 235 cm³ e tubete de polipropileno de 110 cm³ e 50 cm³). Para isso, mudas de cinco meses foram avaliadas quanto à altura da parte aérea, diâmetro do coleto, e número de folhas por planta, bem como Índice de Qualidade de Dickson e área foliar. Mudas de erva-mate produzidas em recipiente biodegradável apresentaram superioridade em altura, diâmetro, número de folhas, área foliar, massa seca da parte aérea e relação entre massa seca da parte aérea e raiz em relação aos tubetes de 100 e 50 cm³ aos cinco meses de cultivo em viveiro. Entretanto, quanto à massa seca da raiz e ao índice de qualidade de Dickson, o cultivo em tubetes de 100 cm³ não foi limitante até os cinco meses. Mudas de erva-mate podem ser cultivadas até os cinco meses tanto em recipientes biodegradáveis quanto em tubetes de 100 cm³. Os recipientes biodegradáveis favorecem o desenvolvimento morfofisiológico de mudas de erva-mate.

Palavras-chave: erva-mate; recipientes biodegradáveis; tubetes.

Abstract

Yerba mate (*Ilex paraguariensis* St. Hill.) is a species with great nutraceutical, pharmaceutical, and food potential, and for this reason its cultivation areas have been expanding, which in turn increases the demand for high-quality seedlings. In this context, this study aimed to compare the growth and development of *Ilex paraguariensis* seedlings produced in three different seedling production units (235 cm³ biodegradable container and 110 cm³ and 50 cm³ polypropylene seedling tubes). To this end, five-month-old seedlings were evaluated for shoot height, root collar diameter, and number of leaves per plant, as well as Dickson Quality Index and leaf area. Yerba mate seedlings produced in biodegradable containers showed superior performance in height, diameter, number of leaves, leaf area, shoot dry mass, and shoot to root dry mass ratio compared with those produced in 100 cm³ and 50 cm³ seedling tubes at five months of nursery cultivation. However, regarding root dry mass and Dickson Quality Index, cultivation in 100 cm³ seedling tubes was not limiting up to five months. Yerba mate seedlings can be cultivated up to five months in both biodegradable containers and 100 cm³ seedling tubes. Biodegradable containers promote the morphophysiological development of yerba mate seedlings.

Keywords: yerba mate; biodegradable containers; seedling tubes.

INTRODUCTION

Yerba mate (*Ilex paraguariensis* St. Hill.) is a tree species native to southern Brazil, Argentina, and Paraguay and belongs to the family Aquifoliaceae. This species has been studied because of its various health benefits, which are due to the production of a wide diversity of secondary metabolites or bioactive compounds (Rodriguez-Mateos *et al*., 2014). The identified compounds include polyphenols, such as chlorogenic acid, and xanthines, such as caffeine and theobromine. These compounds confer notable benefits to human health, including antioxidant, anti-inflammatory, anti-obesity, antidiabetic, neuroprotective, cardioprotective, gut microbiota regulation, and anticancer activities (Cardozo *et al*., 2021).

Although interest in the domestic and international markets has been increasing, commercial plantations in Brazil, located in the southern region of the country, still rely on family labor, have a low level of technification, and show relatively low productivity. With regard to plantation productivity, this is believed to be due to the still incipient stage of genetic improvement programs and the low morphological and physiological quality of the seedlings. Corroborating this hypothesis, Spada *et al*. (2019) states that the productivity of commercial plantations is related to management practices applied in forest seedling production, such as nutrition, irrigation, substrates, and containers, which affect the quality of the seedlings produced.

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For the production of yerba mate seedlings, Wendling *et al.* (2020) recommend the use of plastic seedling tubes, as well as individual biodegradable containers. Seedling tubes have internal ribs that assist in the formation of a well-shaped root system. On the other hand, although they are reusable, this type of container has limitations regarding storage and recycling. For this reason, biodegradable containers have emerged as an alternative capable of replacing nonrenewable plastic materials (Silva, 2011; De Conti *et al.*, 2012), mainly because they allow the formation of a root system that is well distributed in the substrate and capable of growing through the wall of the container in search of resources. However, Wendling *et al*. (2020) emphasize that further evaluations are still required regarding their application on a commercial scale for the production of yerba mate seedlings.

In addition to morphophysiological quality, the commercial production of seedlings involves production cost aspects, which will determine the sale price of the seedlings and often constitute a determining factor in the choice of seedling size. In general, seedling production units that use plastic seedling tubes with lower substrate volumetric capacity support less maximum growth than higher-volume biodegradable containers, which allow a longer nursery period and, in turn, result in larger seedlings and higher final cost. However, little is known about the morphophysiological differences among yerba mate seedlings produced under these conditions. This type of information contributes to the silviculture of the species. Thus, in view of this need, this study aimed to compare the growth and development of *Ilex paraguariensis* seedlings produced in different seedling production units.

MATERIALS AND METHODS

This study was conducted between February and July 2022 at Duffato Commercial Nursery (Monte Castelo, SC) and at Forest Resources Laboratory II of the Federal University of Santa Catarina (Curitibanos, SC). For this study, the term "seedling production units (SPUs)" was used to refer to systems commercially used for producing seedlings of this species and differing in container type and substrate volume.

For seedling production, yerba mate seeds were sown in February 2022 in three types of SPUs: 235 cm³ Ellepot® containers (5 cm in diameter and 12 cm in height) containing a peat-based substrate, and polypropylene seedling tubes with capacities of 110 cm³ (4 cm in diameter and 14.5 cm in height) and 50 cm³ (3 cm in diameter and 12 cm in height), containing a pine bark-based substrate. For base fertilization, 5 g L^{-1} of slow-release NPK fertilizer (15-9-12) was used, and for topdressing, ammonium sulfate (1 g L^{-1}), potassium chloride (2.5 g L^{-1}), and single superphosphate (4.5 g L^{-1}) were used, with six applications being carried out. The seedlings were maintained in a greenhouse until they were five months old.

After the greenhouse development period, the seedlings were measured for shoot height (H), in centimeters, using a graduated ruler, and root collar diameter (D), in millimeters, using a digital caliper with 0.01 mm precision, which allowed the calculation of the H/D ratio (cm/mm). The number of leaves per plant was also counted.

For destructive analysis, the seedlings were removed from their respective containers and washed under running water to remove the substrate. After this procedure, the plants were placed on absorbent paper to remove excess water. Subsequently, the shoot was separated from the root system using pruning shears and weighed on an analytical balance with a precision of 0.01 g. After weighing, the root system and the shoot were individually placed in labeled paper bags and kept in a forced-air oven at 65 °C until constant weight was reached, and then weighed again to determine dry mass. Based on these variables, the Dickson Quality Index (DQI) was calculated according to the methodology of Dickson *et al*. (1960) using the formula $DQI = TDM/\left(\frac{H}{D}\right) + (\frac{SDM}{RDM})$, in which TDM = total dry mass (g); H = shoot height; D = root collar diameter (mm); SDM = shoot dry mass (g); and RDM = root dry mass (g). The shoot dry mass to root dry mass ratio (SDM/RDM) was also estimated.

The leaves of ten seedlings from each container were scanned using an Epson scanner, model V19, to estimate leaf area with the aid of ImageJ software.

The experimental design was completely randomized, with 10 replicates of four seedlings each. The data were evaluated for the assumptions of homogeneity of variances and normality using the Bartlett and Shapiro-Wilk tests at the 5% significance level, respectively. Subsequently, the data were subjected to analysis of variance, and for variables with significant differences, Tukey's test was applied for mean comparison at the 5% significance level. Statistical analyses were performed using R Core Team (2020) software.

RESULTS

A significant difference was observed among the evaluated seedling production units for plant height, root collar diameter, H/D ratio, and number of leaves (Figure 1). Yerba mate plants grown in biodegradable containers showed significantly greater height, diameter, and number of leaves than the other SPUs. For the H/D ratio, no differences were observed between the biodegradable container and the 50 cm³ seedling tube.

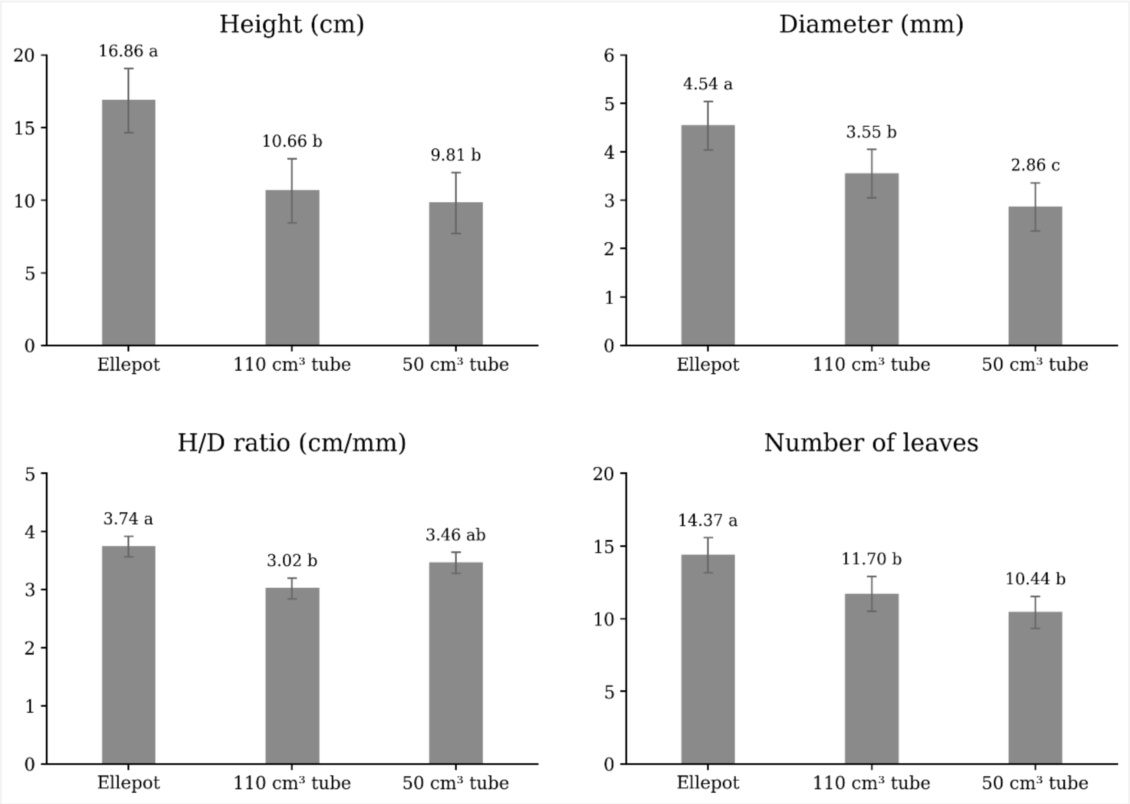

Figure 1 - Height (cm), diameter (mm), height-to-diameter ratio (H/D) and number of leaves of *Ilex paraguariensis* seedlings produced in different containers.

Figura 1 – Altura (cm), diâmetro (mm), relação entre altura e diâmetro (H/D) e número de folhas em mudas de *Ilex paraguariensis* produzidas em diferentes recipientes.

Seedlings produced in biodegradable containers produced 22.8% more leaves than those produced in 100 cm³ seedling tubes and 37.64% more leaves than those produced in 50 cm³ seedling tubes (Figure 1). However, the leaf area of plants produced in biodegradable containers was 162.6% greater than that of seedlings produced in 110 cm³ seedling tubes and 68.64% greater than that of seedlings produced in 50 cm³ seedling tubes (Table 1 and Figure 2).

Table 1 - Leaf area (LA, cm²), shoot dry mass (SDM, g), root dry mass (RDM, g), shoot-to-root dry mass ratio, Dickson Quality Index (DQI), and unit price of *Ilex paraguariensis* seedlings produced in different containers.

Tabela 1 - Área foliar (AF em cm²), massa seca da parte aérea (MSPA em g), massa seca radicular (MSR em g), relação entre massa seca da parte aérea e massa seca radicular, e Índice de qualidade de Dickson (IQD) e valor unitário de mudas de *Ilex paraguariensis* produzidas em diferentes recipientes.

Containers	LA	SDM	RDM	SDM/RDM	DQI	R$
Ellepot	14.52 a*	1.63 a	0.75 a	2.17 a	0.41 a	2.40
110 cm³ tube	5.53 b	0.76 b	0.76 a	1.0 b	0.38 a	1.80
50 cm³ tube	8.61 b	0.68 b	0.53 b	1.28 b	0.24 b	1.50
Mean	9.55	1.02	0.68	1.48	0.34	-
CV (%)	36.79	18.77	28.67	24.12	26.10	-

*Means followed by the same letter do not differ from each other by Tukey's test at a 5% probability of error.
*Médias seguidas de mesma letra não diferem entre si pelo teste de Tukey à 5% de probabilidade de erro.

Seedlings produced in biodegradable containers showed superior shoot development, with a more vigorous appearance and leaves of a more intense green color (Figure 2).

Figure 2 - Five-month-old *Ilex paraguariensis* seedlings produced in different containers. A = Ellepot; B = 110 cm³ tube; C = 50 cm³ tube. Source: authors.

Figura 2 - Mudas de *Ilex paraguariensis* com cinco meses de idade produzidas em diferentes recipientes. A = Ellepot; B = Tubete de 110 cm³; C = Tubete de 50 cm³. Fonte: autores.

DISCUSSION

Cultivation of yerba mate seedlings in the different seedling production units evaluated in this study resulted in significant differences in plant height, root collar diameter, H/D ratio, and number of leaves (Figure 1). In this study, seedlings grown in biodegradable containers showed significantly greater height, diameter, and number of leaves than those produced in the other SPUs. These morphological variables are frequently used to evaluate seedling quality because they are easy to measure and are intuitively understood by seedling producers (Chaves and Paiva, 2004). In addition, these parameters allow seedlings to be classified according to minimum size standards, facilitating decision-making in the nursery and increasing the likelihood of successful forest establishment, which, in turn, depends largely on the morphophysiological quality of the seedlings used (Novaes *et al*., 2014). With regard to seedling height for shipment from the nursery, it is recommended that the height of yerba mate seedlings be considered in relation to container size, and shoot height should be at least 1.5 times the height of the container, in addition to verifying adequate root system formation and the presence of live lateral roots (Wendling *et al*., 2020). For planting, yerba mate seedlings with a minimum height of 12 cm and a root collar diameter of 3 mm are generally recommended (Penteado Junior and Goulart, 2019). Thus, considering height as a criterion, only seedlings produced in biodegradable containers reached the standard for planting after five months of nursery cultivation.

Another aspect observed was that seedlings produced in biodegradable containers produced a greater number of leaves than those produced in 100 cm³ and 50 cm³ seedling tubes (Figure 1). A similar pattern was observed for leaf area, shoot dry mass, and SDM/RDM, which were significantly higher in plants produced in biodegradable containers. However, although the 100 cm³ seedling tube showed an intermediate number of leaves, these were smaller, resulting in reduced leaf area (Table 1 and Figure 2). Leaf area is an important attribute for seedling performance, as the photosynthetic process depends on the interception of light energy and its conversion into chemical energy, with the leaf surface being the basis of the potential yield of a crop (Favarin *et al*., 2002). Under adequate environmental conditions, an increase in leaf area also increases the assimilation area and, consequently, increases production (Silva *et al*., 2007).

Regarding the SDM/RDM ratio, in addition to being significantly higher, only seedlings produced in biodegradable containers reached values greater than 2.0, which is considered ideal (Brissette, 1984). It should be noted that the aboveground portion of the seedlings should not be excessively greater than the root system, as this may impair water absorption and transport to the shoot, especially under field conditions, when water demand is higher for proper initial establishment (Gomes *et al*., 2019).

It is possible that the better development of seedlings grown in biodegradable containers is due to the greater substrate volume (235 cm³), which results in greater availability of nutrients and water. In addition, the main advantages of this type of material include the absence of growth restriction, which allows for better root system architecture, and the possibility of planting the seedling together with the container, without the need for removal, resulting in reduced labor costs. However, seedlings produced in this type of container generally have a higher market price, which may limit their use on a large scale.

Regarding the DQI, yerba mate seedlings reached satisfactory values, which, according to Hunt (1990), should be at least 0.20 for seedlings produced in seedling tubes. Consistent with this, yerba mate seedlings showed satisfactory values when compared with other studies on forest species. In seedlings of *Eucalyptus urophylla*, *Eucalyptus camaldulensis*, and *Corymbia citriodora* at three months of growth, mean DQI values of 0.09 in 50 cm³ seedling tubes and 0.25 in biodegradable containers were reported (Lopes *et al*., 2016). According to some authors, the Dickson Quality Index is a reliable indicator of seedling quality, as it considers both robustness and the balance of biomass distribution, integrating important variables used in seedling evaluation (Dickson *et al.*; Lima Filho *et al*., 2019).

It is worth noting that the evaluated seedlings were five months old, indicating that up to this stage the 110 cm³ seedling tube was not a limiting factor for the growth of yerba mate seedlings for these variables. From an economic perspective, this seedling costs the consumer R$ 1.80 per unit, whereas seedlings produced in biodegradable containers cost R$ 2.40 per unit, and, considering that no differences in growth were observed, the use of seedlings produced in 110 cm³ seedling tubes may result in cost savings for the consumer. Seedlings produced in biodegradable containers and those produced in 110 cm³ seedling tubes showed very similar root dry mass values, which was not expected given the more robust appearance of the root system in seedlings produced in biodegradable containers. In addition, seedlings produced in biodegradable containers showed superior shoot development, with a more vigorous appearance and leaves of a more intense green color (Figure 2). It is believed that the superior appearance of the root system developed in biodegradable containers is due to its better conformation, which is related to the abundance of fine roots distributed throughout the substrate volume and their aggregation, promoting greater root ball integrity (Delgado *et al*., 2017), thus justifying the higher investment in these seedlings.

Based on the observed results, yerba mate seedlings produced in biodegradable containers showed greater height, diameter, number of leaves, leaf area, shoot dry mass, and the ratio between shoot dry mass and root dry mass than those produced in 100 and 50 cm³ seedling tubes after five months of nursery cultivation. However, with regard to root dry mass and the Dickson Quality Index, cultivation in 100 cm³ seedling tubes was not limiting up to this stage, and further studies with seedlings older than five months are recommended.

CONCLUSIONS

- Yerba mate seedlings can be cultivated up to five months in both biodegradable containers and 100 cm³ seedling tubes.
- Biodegradable containers promote the morphophysiological development of yerba mate seedlings.

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