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Organic substrates in the development of camu-camuzeiro *(Myrciaria dubia (H. B. K.) McVaugh)* in the amazon region

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Abstract

Amazon is the largest tropical forest on the planet, it has a variety of plant species with emphasis on many fruit trees, such as the camu-camuzeiro (*Myrciaria dubia* (H. B. K.) McVaugh), which occurs naturally on the banks of rivers, lakes, lowlands and flooded forest of the Amazon. The objective of this study is to evaluate the development of camu-camuzeiro seedlings in different organic substrates. The experiment was conducted in the seedling production nursery of the Federal Rural University of Amazon. The experimental design was entirely randomized, with ten treatments and five repetitions, each portion was represented by ten plants, totaling 500 seedlings. The substrates evaluated were: T1 - Humus; T2 - Humus + Bird manure; T3 - Humus + Bovine manure; T4 - Humus + Açaí kernel; T5 - Humus + Chestnut shell; T6 - Humus + Chestnut shell + Poultry manure; T7 - Humus + Bovine manure + Chestnut shell; T8 - Humus + Açaí kernel + Poultry manure; T9 - Humus + Açaí kernel + Bovine manure; T10 - Commercial. The different substrates used influenced the development of camu-camuzeiro seedlings evaluated at 180 days. The treatments with a substrate based on Humus (T1), Humus + Açaí kernel (T4), Humus + Chestnut shell (T5), Humus + Chestnut shell + Poultry manure (T6), Humus + Bovine manure + Chestnut shell (T7), Humus + Açaí Kernel + Bovine manure (T9) and the commercial substrate (T10) are the best for a satisfactory development of this crop in the production field.

Keywords: Amazon, biodiversity, fruticulture, myrtaceae, production

Introduction

The Amazon biome has a hydrographic immensity, and plant and animal biodiversity, which makes it one of the largest hosts of existent species on Earth. Thus, Amazon is known as the largest rainforest on the planet, and all this vast biodiversity holds 1/3 of all forest reserves of the humid tropics and grows on its territory to more than 30 thousand plant species (IBAMA, 2017). In this diversity of plant species, the numerous fruit trees stand out, as the camu-camuzeiro (*Myrciaria dubia* (H.B.K.) McVaugh) which is typically Amazonian (Yuyama, 2011).

The camu-camuzeiro is a fruit-bearing species, also known as "araçá-d'água" and "caçari" depending on the region, which occurs naturally on the banks of rivers, lakes, lowlands and flooded forest of the Amazon, from the central region of the state of Pará to the region of Peru, Colombia and Venezuela (Teixeira et al., 2004). One of the particularities of camu-camuzeiro is related to its fruits and its levels of ascorbic acid that can reach 7.355 mg 100 g $^{-1}$ of pulp, which has aroused consumer market interest (Chagas et al., 2012).

According to Pinedo Panduro et al. (2018), this species has a direct relationship with floristic parameters that span the whole Amazonian region where it can be found, and this factor has been essential to Camu-camuzeiro to be considered the first species of both economic and ecological importance of the Amazon biodiversity that develops in floodable ecosystems.

The peculiarities of camu-camu, the fruit of camu-camuzeiro, has provided food with excellent supplement rich in vitamin C, through products such as concentrated drinks, vitamins, nectar, carbonated soda, jelly, ice cream, chocolates. As for cosmetics, it can be used in shampoo, conditioner, and facial creams. In addition to these utilities, it can also be manipulated for

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manufacturing in the drug network in the form of pills, tablets, and micro capsulated (Yuyama, 2011).

It is worth emphasizing that this fruit-bearing species occurs in flooded areas and that this causes different levels of flooding due to the growing site edaphoclimatic characteristics, which facilitates the loss of these nutrients. The absence of low-cost fertilization technologies does not allow for the supply of nutrients in adequate quantities to the species to restore it year after year during its productivity cycle (Pinedo Panduro et al., 2018).

This requires the use of organic fertilization techniques based on the incorporation and applicability of organic compounds that are formed by plant and animal residues in soils and substrates adhered to the plant (Abanto-Rodriguez, 2019). Given this context, this study has the objective of evaluating the development of seedlings of camu-camuzeiro in different organic substrates, through the use of plant and animal residues.

Materials and Methods

The experiment was conducted at the seedling production nursery of the Federal Rural University of Amazon, located in the municipality of Belém, Pará, with geographic coordinates 1°27'19.9"S 48°26'18.6"W. The Köppen climatic classification is Af, which classifies it as always humid, in such manner that, the seasonality of the rainy season occurs in the months of December, January, February, March, April, and May. The fruits utilized in this research were harvested in the complete maturation stage in the Active Germplasm Bank of Embrapa Amazônia Oriental, whose clone was the CPTU 6, due to it presenting the largest fruits and the higher pulp yield, in addition to it being the most productive. After harvesting, the fruits were immersed in water for approximately twenty-four hours, then pulped manually and the pulp residues were removed.

The camu-camu seeds were put to germinate in plastic trays, containing as a substrate organic compound + vermiculite in a 2:1 ratio, for a period of 30 days. Following this period, the most vigorous seedlings in the "match stick" stage (when the first leaf is still closed) were selected and transplanted to the 0,299 dm^3 tubes, where different types of substrates were utilized in a 3:1 and 3:1:1 ratio. Irrigation was carried out daily, through micro-sprinklers distributed on-site.

The experimental design was entirely randomized, with ten treatments and five repetitions, each portion was represented by ten plants, totaling 500 seedlings. The substrates evaluated for the production of camu-camuzeiro seedlings were: T1 - Humus; T2 - Humus + Bird manure; T3 - Humus + Bovine manure; T4 - Humus + Açaí kernel; T5 - Humus + Chestnut shell; T6 - Humus + Chestnut shell + Poultry manure; T7 - Humus + Bovine manure + Chestnut shell; T8 - Humus + Açaí kernel + Poultry manure; T9 - Humus + Açaí kernel + Bovine manure; T10 - Commercial (which has in its composition pine bark, coconut fiber, potassium nitrate, and simple superphosphate).

The humus analysis was conducted in the soil laboratory of the Federal Rural University of Amazon, while the other compounds were utilized according to the orientation of the studied literature for the discussion of this work. The chemical characteristics of the organic compounds evaluated are shown in (**Table 1**).

Table 1. Table of chemical characteristics of poultry litter, bovine manure, açaí kernel, and chestnut shell, used as organic substrates

Organic substrates	pH	N	P	K	Ca
		gKg^{-1}			$Cmolc/dm^{-3}$
Humus	3.7	-	20.5	0.05	1.9
Poultry manure	-	26.3	27	38	-
Bovine manure	7.7	12	2.7	5.1	13
Açaí kernel		5.97	2.15	8.13	1.69
Chestnut shell		4.4	2.18	3.3	16.01

Source: Soares, 2014; Elacher, 2014.

During the development period, the camu-camuzeiro seedlings were in a nursery with a shade screen with 50% light and rain interception, for a period of six months, being evaluated monthly. At the end of 180 days according to the methodology adapted from Delarmelina et al. (2014), the following evaluations were performed: Plant Height (PH), expressed in centimeters with the aid of a ruler; total Number of Leaves (NOL), the count of all existing leaves in the seedlings; and Collar Diameter (CD), expressed in millimeters with the aid of digital caliper.

In the destructive morphometric analysis, five samples of each treatment were collected in all repetitions, totaling 25 samples per treatment, taken randomly, for evaluation of the following variables: Root Length (RL), to determine the RL the seedlings were taken from the tubes so that the roots could be cleaned with running water. Subsequently, the seedlings were put to dry in the shade for about an hour, being separated by the cutting at the collar base, the measurements were expressed in centimeters measured with the help of a ruler.

To determine the variables Fresh Mass of Shoot part (FMS), Fresh Mass of Roots (FMR), Dry Mass of Shoot part (DMS), and Dry Mass of Roots (DMR), these were weighed on digital scales. To determine both the DMS and DMR, the samples were placed in paper bags with identification, dried in greenhouses at 60°C, for 48 hours, and then weighed again to determine dry mass.

Absolute growth rate data of height AGRH (Equation 1) and seedling diameter AGRD (Equation 2) were obtained through the data collected at the end of the experiment. According to Barbieri Junior et al. (2007), the absolute growth rate (AGRH) is determined by the growth variation in grams per day between two consecutive evaluations throughout the cycle. Therefore, it was calculated as follows:

$$AGRH = \frac{H_2 - H_1}{T_2 - T_1} \text{ (cm/day)} \quad \text{(Equation1)}$$

Where: AGRH = Absolute growth rate of seedlings height; H_1 and H_2 = Actual height values measured in two different periods; and T_1 and T_2 = Period (in days) where the evaluations were carried out.

$$AGRD = \frac{D_2 - D_1}{T_2 - T_1} \text{ (mm/day)} \quad \text{(Equation 2)}$$

Where: AGRD = Absolute growth rate of seedlings diameter; D_1 and D_2 = Actual diameter values measured in two different periods; and T_1 and T_2 **=** Period (in days) where the evaluations were carried out.

After the weighing of the shoot part and the root, the mean of each treatment was analyzed using the Dickson index quality (DIQ) (Equation 3) (Dickson et al., 1960), which consists of the following formula:

$$IQ = \frac{TDM}{[SPH(cm)/CD(mm)]+[SPDM(g)/RDM(g)]} \quad \text{(Equation 3)}$$

Where: TDM = Total dry mass (g); SPH = Shoot-part height (cm); CD = Collar diameter (mm); SPDM = Shoot-part dry mass (g) and RDM = Root dry mass (g).

To study the effects of different substrates on the evaluated variables, initially, the data were submitted to the Levene test to assess the variance homogeneity, followed then by the Kolmogorov-Smirnov test to appraise the normality, for the variables in which these assumptions were not met, their respective values were then transformed into $(x + 0,5)^{0,5}$. Thus, after verifying that such assumptions were met in all variables, the application of variance analysis (ANOVA) was performed.

When significant differences in Anova were identified, the Scott-Knott test was applied at a 5% probability level. The statistical procedures were carried out with the aid of AgroEstat software (2015) (Barbosa & Maldonado, 2015).

Results and Discussion

The height variation, collar diameter, number of leaves, root length, and shoot length analyses showed that there was a significant influence between substrates utilized in the production of camu-camuzeiro seedlings based on the F test ($P < 0.05$).

The different utilized substrates for the production of camu-camuzeiro seedlings differed significantly in height, diameter, the number of leaves, root length, and shoot length. For plant height (**Figure 1**A) a maximum value of 37.6 cm was observed in the T4 treatment (Humus + Açaí kernel), however, the T2, T3, T6, T7, T8, and T9 treatments statistically resembled the T4 (Figure 1A).

Regarding the collar diameter, the treatment with the best mean for seedlings development was the T7 (Humus + Chestnut shell + Bovine manure) with 3.14 mm (Figure 1B).

Concerning the number of leaves, statistical differences were observed between the substrates used. The best means for this variable, which had statistically similar results were the T4 (Humus + Açaí kernel), T6 (Humus + Chestnut shell + Poultry manure), T7 (Humus + Chestnut shell + Bovine manure), and T8 (Humus + Açaí kernel + Poultry manure) treatments with an average number of leaves of 50.9 (Figure 1C).

The best means for the seedlings root length were from the treatments with Humus (T1), Humus + Poultry manure (T2), Humus + Chestnut shell (T5), Humus + Bovine manure + Chestnut shell (T7), Humus + Açaí kernel + Poultry manure (T8), Humus + Açaí kernel + Bovine manure (T9) and the Commercial one (T10) (Figure 1D).

Souza et al., (2017), studied the influence of seed size on the vigor of seedlings of camu-camuzeiro populations cultivated in the commercial substrate, where they identified the mean height of 23.9 cm, a diameter of 2.17 mm, and a mean number of leaves of 34.3, these results are below those found in this work for the same parameters, in which the highest mean for the variable collar diameter (3.14 mm) was for the seedlings of the substrate in T7 (Figure 1B).

Gomes et al. (2003), cite that the seedling height, collar diameter, and root length are parameters that express the quality of the seedling. Nevertheless, the collar diameter, evaluated separately or combined with the height, is considered one of the best morphological characteristics to conjecture seedling quality standards. These should have larger collar diameters to obtain a better balance of growth of the shoot-part (Gomes et al., 2015).

In the following variables, height, collar diameter, and leaf emission, the best means were found in treatments compounded by bovine and poultry manure (Figure 1A, B, and C), a possible explanation for this result may be the fact that in these substrates components are in a favorable combination to the seedlings development. It should be emphasized that these three variables are important

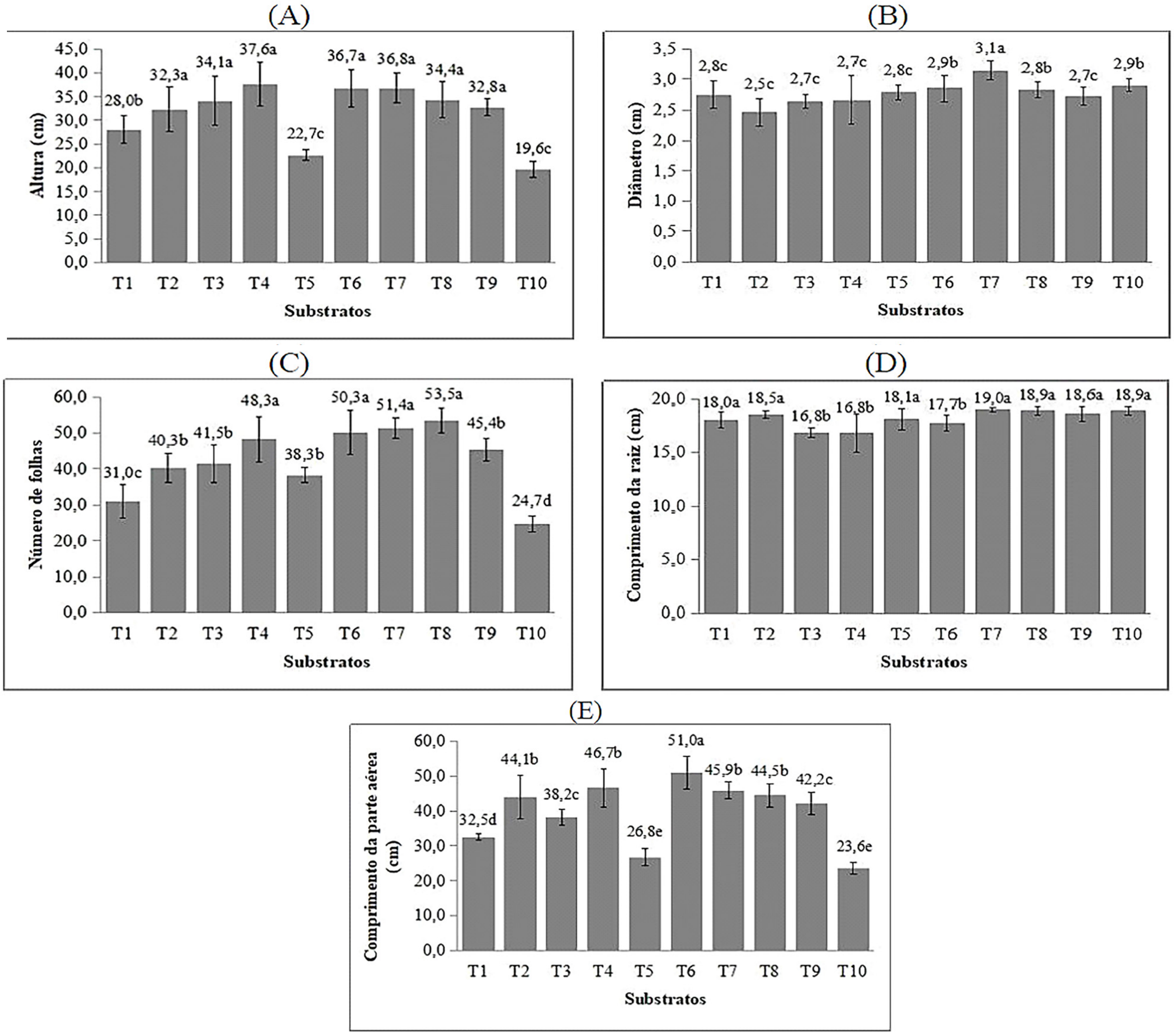

Figure 1. Mean ± standard error for variable height (a), diameter (B), number of leaves (C), root length (D), and shoot length (E) of *Mr. dubia* seedlings on different substrates. Means followed by equal letters do not differ from each other using the Scott-Knott test at 5% probability of error.

in the development to compose a commercialization pattern, since the camu-camuzeiro does not yet have a pattern registered in the Ministry of Agriculture, Livestock, and Supply (MAPA), for being an Amazonian native species in the process of domestication.

According to Faria et al. (2016), the N, P, and Mg contents present in poultry and bovine manure along with the physical properties of total pore volume and bulk density contribute to seedling growth. Vieira et al. (2015), state that there is a relationship of nitrogen and potassium that makes it possible to increase the chlorophyll content in the leaves, this content connects directly with the production of the dry mass of the shoot and with the accumulation of nitrogen in the leaves. It is noteworthy that according to Malavolta (1980), nitrogen is the main element in leaf emission, and the lack of this element compromises seedling growth and reduces the accumulation of dry matter.

Sousa et al. (2010), point up that all seedlings produced should have a good development of its root system and that it should be well-formed for it to be efficiently transplanted to a definitive place, which will allow for the best fixation to the soil and better absorption of water and nutrients, thus presenting a faster and more efficient development.

For the characteristics variance analysis: fresh mass of root and fresh mass of shoot part had a significant difference ($P < 0,05$) between the different substrates utilized in camu-camuzeiro seedlings production (**Figure 2**).

The highest mean of the fresh mass of root was obtained from the seedlings that developed in the control substrates Humus (T1) and the Commercial (Plantimax®) (T10) (Figure 2A). For the fresh mass of shoot part, the

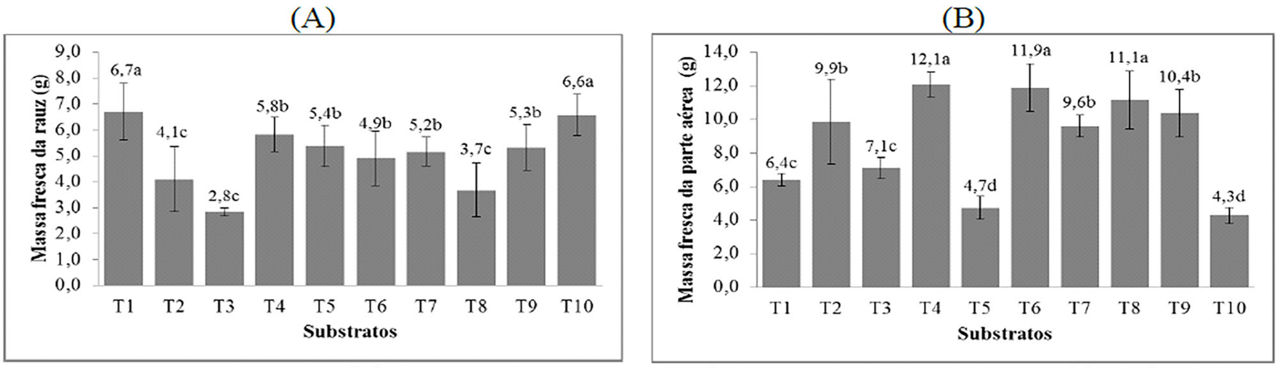

Figure 2. Mean ± standard error for the fresh mass of root (FMR) (A) and fresh mass of shoot part (FMS) (B) of *Myrciaria dubia* seedlings on different substrates. Means followed by equal letters do not differ from each other using the Scott-Knott Test at 5% probability of error.

Humus + Açaí kernel (T4), Humus + Chestnut shell + Poultry manure (T6), and Humus + Açaí kernel + Poultry manure (T8) had the highest means (Figure 2B).

There was a statistical difference in the parameters variance analysis: dry mass of root and dry mass of shoot part between the treatments with different substrates used in the production of *M. dúbia* seedlings (**Figure 3**).

The evaluation of the dry mass of the root that developed in the substrates Humus (T1), Humus + Açaí kernel (T4), Humus + Chestnut shell (T5), Humus + Bovine manure + Chestnut shell (T7), and Commercial (10), presented significant higher gains compared to that grown in the substrate Humus + Bovine manure (T3), that obtained the lowest mean (Figure 3A).

Such a fact can be explained due to the necessity of oxygen for the seedling roots respiration, which occurs from the substrate itself, therefore the substrate must have good aeration to promote greater root and leaf area growth (Sasso et al., 2010). For Marques et al. (2017), the substrate besides being essential, assists in root development, being responsible for a satisfactory formation of seedlings, making them viable for planting.

It was possible to observe that the dry matter weight of the shoot was inversely proportional to the weight of the dry mass of root for most treatments since the highest averages were from the seedlings of: Humus + Açaí kernel (T4), Humus + Chestnut shell + Poultry manure (T6), Humus + Açaí kernel + Poultry manure (T8) and Humus + Açaí kernel + Bovine manure (T9) (Figure 3B).

When relating production of fresh and dry mass of shoot part, it was noted that there was a direct relationship, where Humus + Açaí kernel (T4), Humus + Chestnut shell + Poultry manure (T6), Humus + Açaí kernel + Poultry manure (T8) and Humus + Açaí kernel + Bovine manure (T9) did not differ from each other (Figure 3B). Barros et al. (2020), when evaluating the effect of different organic substrates on Papaya seedlings, found that the corral manure provided the best development of the seedlings. Other authors also emphasize the importance of animal manure in the formulation of substrates for the production of seedlings as they promote the largest number of leaves and associate this effect with the increase of organic matter in these substrates (Cavalcante et al., 2016; Pinheiro et al., 2018; Guse et al., 2021).

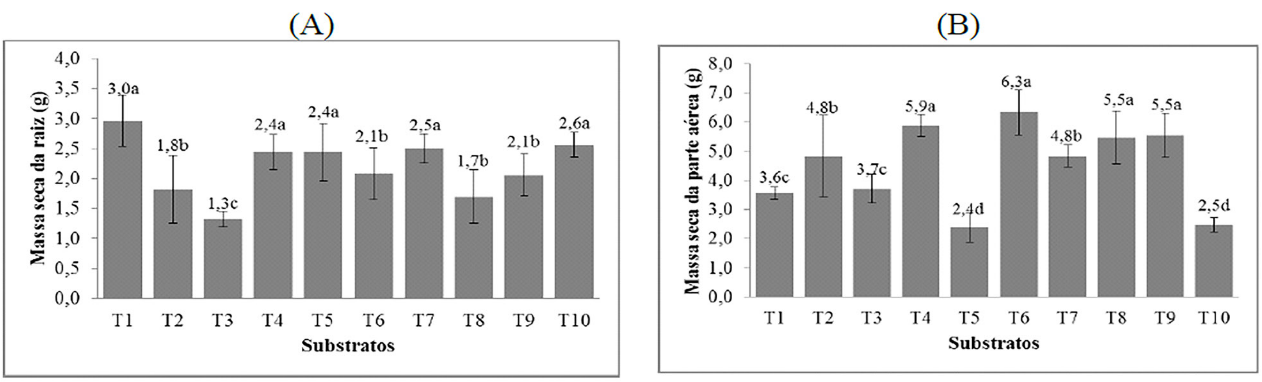

Figure 3. Mean ± standard error for the fresh mass of root (FMR) (A) and fresh mass of shoot part (FMS) (B) of *M. dubia* seedlings on different substrates. Means followed by equal letters do not differ from each other using the Scott-Knott Test at 5% probability of error.

According to Pelloso (2011), the weight of the dry matter of the roots is a parameter that represents the level of survival and growth of seedlings infield, that is, the higher and more abundant the root system, regardless of the height of the shoot-part, this will indicate whether the plant can withstand the field conditions at the time of transplantation to a definite place. There is a close relationship between the dry matter weight of the shoot-part and the dry matter weight of the roots.

The parameters variance analysis, the absolute growth rate of height, and Dickson quality index were significant in the F test (P < 0.05) between the different substrates for camu-camuzeiro seedlings production, however, there was no significant difference for the absolute growth rate of diameter.

For the absolute growth of height (**Figure 4**A), it can be observed that the seedlings cultivated in the commercial substrate (T10), were significantly lower in average, followed by the seedlings cultivated in the Humus substrate (T1) and Humus + Chestnut shell substrate (T5), the others treatments presented the highest AGRH, while did not differ statistically, with the range of variation of 0.16 to 0.20 cm/day, with an AGRH average of 0.174 cm/day. While for the seedlings absolute growth rate of diameter there was no significant difference between the various substrates, presenting an average of 0,011 mm/day (Figure 4B). Regarding the Dickson quality index (DQI), seedlings cultivated in the commercial substrate (T10) obtained the highest mean, followed by the seedlings of treatments with Humus (T1), Humus + Açaí kernel (T4), Humus + Chestnut shell (T5), Humus + Chestnut shell + Poultry manure (T6), Humus + Bovine manure + Chestnut shell (T7), Humus + Açaí Kernel + Bovine manure (T9) (Figure 4C).

Santos et al. (2010), point out that the absolute growth rate indicates the growth rate of the plant under physiological aspects, however, it is more admissible to define growth rate according to the plant weight. For seedlings from treatments with low development, it may be due to a lower nutrients absorption, this means there is difficulty in releasing important minerals, and therefore lower investment of the plant in its development (Brito et al., 2018).

Regarding DQI, this index is traditionally used in the production of forests species seedlings, but the same characteristics that compose the DQI are important for the production of fruit seedlings. (Dias et al., 2012). In addition, it is relevant in the initial in-field growth evaluation of fruit trees (Abanto-Rodríguez et al., 2016). The higher the value of such index, the higher the seedling quality standard (Gomes et al., 2003).

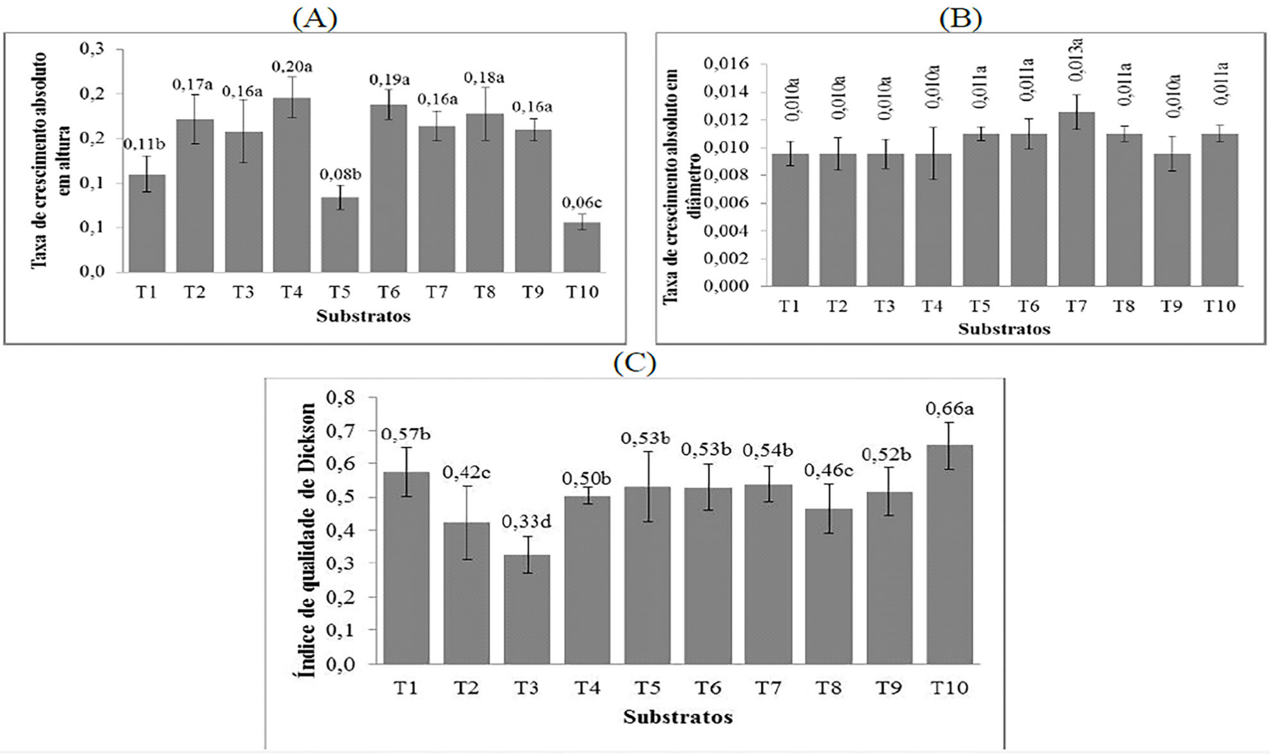

Figure 4. Mean for absolute growth rate in height (A), the absolute growth rate in diameter (B), and the Dickson quality index (DQI) (C) of seedlings of *Mr. dubia* on different substrates. Means followed by equal letters do not differ from each other using the Scott-Knott Test at 5% probability of error.

Conclusion

Camu-camuzeiro seedlings development is influenced by the use of different substrates, according to the data presented in this work, it is recommended that the Humus (T1), Humus + Açaí kernel (T4), Humus + Chestnut shell (T5), Humus + Chestnut shell + Poultry manure (T6), Humus + Bovine manure + Chestnut shell (T7), Humus + Açaí Kernel + Bovine manure (T9) and the commercial substrate (T10) are the best for a satisfactory development of camu-camuzeiro seedlings in the production field. It would be interesting for future work to conduct tests with each individualized substrate.

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