

Crop Science/ Original Article

#### ISSN 1678-3921

Journal homepage: www.embrapa.br/pab

For manuscript submission and journal contents, access: www.scielo.br/pab

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Received October 03, 2023

Accepted January 22, 2024

#### How to cite

BALBINO, T.J.; ESPINDULA, M.C.; TAVELLA, L.B.; TEIXEIRA, R.G.P.; BRAVIN, N.P.; DIAS, J.R.M.; BRAVIN, M.P. Alternative substrates for the production of clonal *Coffea canephora* seedlings. **Pesquisa Agropecuária Brasileira**, v.59, e03538, 2024. DOI: https://doi.org/10.1590/S1678-3921.pab2024.v59.03538.

# Alternative substrates for the production of clonal *Coffea* canephora seedlings

**Abstract** – The objective of this work was to evaluate the physicochemical properties of coffee husk, elephant grass, and sugarcane alone or combined with each other or with commercial compounds, as well as their use as substrate for the production of clonal Coffea canephora seedlings. The experiment was carried out in two stages: one for the physicochemical characterization of the substrates, and the other for evaluations of the growth of coffee seedlings in these substrates. In the first stage, the treatments consisted of coffee husk (CH), elephant grass (EG), sugarcane (SC), commercial substrate (CS), vermiculite (VM), and their combinations. In the second stage, the standard substrate (soil) was also evaluated. CH presented a high electrical conductivity and density and a low aeration space and available water. SC stood out for its lower pH and density and its greater porosity. SC and EG were viable for coffee seedling production, not differing from the commercial and standard substrates. With the exception of CH+EG, CH+CS, and EG+SC, the combined substrates allowed of a greater seedling growth than the isolated ones. CH was only viable as a substrate when combined, especially with VM. Combining CH, SC, and EG to the standard substrate improves the quality of clonal C. canephora seedlings.

**Index terms**: coffee, organic waste, physicochemical property, tubes, vegetative propagation.

## Substratos alternativos para produção de mudas clonais de *Coffea canephora*

Resumo - O objetivo deste trabalho foi avaliar as propriedades físicoquímicas da palha de café, do capim-elefante e da cana-de-açúcar isolados ou combinados entre si ou com compostos comerciais, bem como seu uso como substrato para a produção de mudas clonais de Coffea canephora. O experimento foi conduzido em duas etapas: uma para a caracterização físicoquímica dos substratos, e a outra para a avaliação do crescimento das mudas de café nesses substratos. Na primeira etapa, os tratamentos consistiram de palha de café (PC), capim-elefante (CE), cana-de-açúcar (CA), substrato comercial (SC), vermiculita (VM) e suas combinações. Na segunda etapa, avaliou-se, também, o substrato padrão (solo). A PC apresentou altas condutividade elétrica e densidade e baixos espaço de aeração e água disponível. A CA se destacou por seus menores pH e densidade e sua maior porosidade. A CA e o CE foram viáveis para a produção de mudas de café, sem diferir dos substratos comercial e padrão. Com exceção de PC+CE, PC+SC e CE+CA, os substratos combinados proporcionaram maior crescimento das mudas que os isolados. A PC foi viável como substrato apenas em combinação, principalmente com VM. A combinação de PC, CA e CE com o substrato padrão melhora a qualidade de mudas clonais de C. canephora.

**Termos para indexação**: café, resíduo orgânico, propriedades físico-químicas, tubetes, propagação vegetativa.



#### Introduction

Coffee is the second most consumed beverage worldwide, with Brazil ranking first as the main coffee producer and exporter (ICO, 2023). In the country, the species *Coffea canephora* Pierre ex A.Froehner represents 30% of the total coffee production, occupying a planted area of around 388 thousand hectares (Acompanhamento..., 2023). In this scenario, the development of new coffee crops and the renewal of the coffee park in Brazil, mainly in *C. canephora*-producing areas, have increased demands for clonal *C. canephora* seedlings, whose quality is crucial for the successful enterprise of perennial crops, such as coffee (Guisolfi et al., 2020).

A quality coffee seedling must present a vigorous development and a well-formed root system (Marana et al., 2008; Pereira et al., 2013), both of which depend on genetic characteristics and the interaction of the plant material with the environment (Dardengo et al., 2013). In the second case, the used substrate and container are factors that will directly influence the quality of the seedlings (Klein, 2015; Meneghelli et al., 2018).

In Brazil, currently, the main production systems for coffee seedlings are plastic bags (conventional) and polyethylene tubes (Guisolfi et al., 2020). In the conventional system, the standard substrate recommended is a mixture of 70 to 80% subsurface soil and 20 to 30% organic fertilizer (Espindula et al., 2015). However, high substrate volumes are required to fill the containers and an inappropriate use of these substrates can compromise the sustainability of coffee production and pose biological risks due to contamination by pathogens, especially nematodes (Montagne et al., 2016).

In the production system in tubes, commercial substrates are generally used in smaller amounts, reducing the risk of contamination, problems with root curling, and the space required in the nursery and for transport (Espindula et al., 2015; Guisolfi et al., 2020). However, although commercial substrates are efficient for seedling growth, their high costs may be unviable for family farmers. An alternative is using organic waste in the formulation of the substrate to reduce production costs in this system, decreasing the amount of or even replacing the used commercial substrate (Kratz et al., 2013; Siqueira et al., 2020).

Several materials have been used as alternative substrates for *C. canephora* seedling production,

such as processed wood residue (Aquino et al., 2017), residues of pepper powder, coconut fiber, pine bark (Guisolfi et al., 2020), coffee husk, (Meneghelli et al., 2018), cattle manure (Silva et al., 2013), and tannery sludge (Berilli et al., 2014). However, some of these substrates may not be accessible in all coffeeproducing regions, being abundant only in certain areas. For example, the coffee husk residue, rich in potassium and nitrogen, is a byproduct of coffee grain processing that can be easily obtained in the state of Rondônia, Brazil, where coffee farming is practiced, generating a 1:1 grain-to-husk ratio (Silva et al., 2020). Another material found in the region is elephant grass (Pennisetum purpureum Schumach.), an important forage species widely used for cutting due to its high dry matter production capacity and good adaptation to most tropical ecosystems (Leal et al., 2013). In addition, sugarcane (Saccharum spp.) appears to be a promising material for formulating substrates because it is a widely available residue and has a high biomass productivity (Klein, 2015).

An ideal substrate should be able to support plant cuttings and provide sufficient amounts of water and nutrients, as well as be easily accessible, economically viable, and free of pests, phytopathogens, and foreign seeds, also showing a low density, good porosity, and water retention capacity under low humidity tensions, in addition to nutritional balance (De Boodt & Verdonck, 1972; Kratz et al., 2013; Zorzeto et al., 2014; Klein, 2015).

Therefore, the characterization and understanding of a substrate's physical and chemical properties are essential to ensure quality products. Physical characteristics, in particular, are especially important, as substrate air-water ratios should not be altered during cultivation (Schafer et al., 2015). Klein (2015) and Schafer et al. (2015) highlighted that the characteristics of the substrate vary according to its composition, origin, and form of raw material production, which can lead to different seedling-development results.

The objective of this work was to evaluate the physicochemical properties of coffee husk, elephant grass, and sugarcane alone or combined with each other or with commercial compounds, as well as their use as substrate for the production of clonal *C. canephora* seedlings.

#### **Materials and Methods**

The research was carried out at the experimental field of Embrapa Rondônia, located in the municipality of Ouro Preto do Oeste, in the state of Rondônia, Brazil (62°15'10"W, 10°45'43"S, at 300 m above sea level). The predominant climate in the region is Am, tropical rainy, according to Köppen's classification, with an average annual temperature of 25°C and average rainfall rates from 1,900 to 2,200 mm per year (Alvares et al., 2013).

The experiment was carried out in two stages: in the first, the physicochemical characterization of the substrates was performed; and, in the second, the potential of the substrates to produce seedlings was evaluated. The used substrates were: coffee husks of *C. canephora* fruits; biomass from the Cameroon elephant grass cultivar; biomass from the CB47-355 (Mulata Pelada) sugarcane variety; the Agrofloc expanded vermiculite (Brasil Minérios, Goiânia, GO, Brazil); and the Tropstrato HT commercial substrate, composed of peat, expanded vermiculite, and pine bark (Vida Verde, Mirim Mogi, SP, Brazil) (Table 1).

The coffee husks were obtained at the coffee processing unit of the experimental field of Embrapa Rondônia, located in the municipality of Ouro Preto do Oeste, in the state of Rondônia, Brazil. The biomasses of elephant grass and sugarcane were obtained at the experimental field of Embrapa Rondônia, located in the municipality of Presidente Médici, also in the state of Rondônia. Both elephant grass and sugarcane were harvested manually (entire aerial part of the plants), keeping two nodes above the ground for elephant grass plants, which had been regrowing for four months after the last cut, and a height of 20 cm for sugarcane, enabling the regrowth of the ratoons and the recompositing of the plants.

In the first stage of the experiment, the experimental design was a randomized complete block, with the following 20 treatments for the analysis of the substrates alone or combined (T1–T20), with three replicates: T1, composted coffee husk (CH); T2, composted elephant grass (EG); T3, composted sugarcane (SC); T4, commercial substrate (CS); T5, expanded vermiculite (VM); T6, CH+EG at a 1:1 ratio; T7, CH+SC at a 1:1 ratio; T8, CH+CS at a 1:1 ratio; T9, CH+VM at a 1:1 ratio; T10, EG+SC at a 1:1 ratio; T11, EG+CS at a 1:1 ratio; T12, EG+VM at a 1:1 ratio; T13, SC+CS at a 1:1 ratio; T14, SC+VM at a 1:1 ratio; T15, CH+SC+EG at a 1:1:1 ratio; T16, CH+SC+EG+CS at a 1:1:1:1 ratio; T17, CH+SC+EG+CS+VM at a 1:1:1:1:1 ratio; T18, CH+SC+VM at a 1:1:1 ratio; T19, CH+EG+VM at a 1:1:1; and T20, CH+EG+SC+VM at a 1:1:1:1.

Substrate composting (biostabilization) conducted in a greenhouse from April to June 2015. The compost pile was built with layers approximately 10 cm thick. Between the layers, 2.2 kg triple superphosphate were evenly distributed for each cubic square of material, following the recommendations of Souza & Resende (2006), with the application of 8.16, 6.60, and 8.66 g kg<sup>-1</sup> urea on the biomasses of coffee straw, elephant grass, and sugarcane, respectively. Urea was applied according to Gomes et al. (2001), considering the following percentages of carbon and nitrogen contained in each raw material: 51 and 0.62% in coffee husk (Souza & Resende, 2006), 40 and 0.46% in sugarcane (Trivelin et al., 1997), and 53 and 0.62% in elephant grass (Leal et al., 2013). Temperature variations in the piles were monitored daily with a digital infrared thermometer. The compounds were considered ready for use when their internal temperature became stable, at a close-to-room

**Table 1.** Chemical characteristics of the raw materials and commercial substrates used for the production of clonal *Coffea canephora* seedlings<sup>(1)</sup>.

Raw material and commercial substrate	N	P	K	Ca	Mg	S		
	(g kg <sup>-1</sup> )							
Coffee husk	35.0	1.1	39.0	17.1	1.8	1.4		
Elephant grass	25.0	10.8	16.0	14.6	2.0	1.0		
Sugarcane	25.0	15.0	21.0	20.9	1.8	0.9		
Commercial substrate	4.0	2.1	5.0	14.0	11.4	1.0		
Vermiculite	_	0.5	22	1.3	126.8	_		

<sup>(1)</sup>N, obtained by the Kjeldahl method; and P, K, Ca, Mg, and S, determined in acid extract (nitric and perchloric acid).

temperature of 30±2°C, at about 100 days after the beginning of the biostabilization process.

For their physicochemical characterization, samples of the substrates were sent to the Laboratory of Plant Substrates of the Department of Horticulture and Forestry of Faculdade de Agronomia of Universidade Federal do Rio Grande do Sul. Electrical conductivity and hydrogen potential (pH) were evaluated at a 1:5 dilution (v:v) in accordance with Instrução Normativa SDA no. 17, de 21 de maio de 2007 (Brasil, 2007), detailing aspects of the law on the standards for plant substrate analysis. Dry density was determined using the self-compaction method (Hofmann, 1970). Water retention curves at 0, 10, 50, and 100 hP, obtained according to De Boodt & Verdonck (1972), were prepared to determine total porosity at 0 hPa, aeration space at 10 hPa, available water at 10 to 100 hPa, readily available water (RAW) at 10 to 50 hPa, buffering water at 50 to 100 hPa, and remaining water at 100 hPa.

In the second stage, the performance of the substrates in the formation of *C. canephora* clonal seedlings was evaluated in a greenhouse from September to December 2015. The treatments consisted of the 20 substrates previously described, but placed in 280 cm<sup>-3</sup> tubes, plus a standard substrate (T21), i.e., subsurface soil, packaged in 700 cm<sup>-3</sup> polyethylene bags. The experimental design was randomized complete blocks with four replicates and one cutting per replicate.

In T21, the used soil was an eutrophic Ferralsol, with the following chemical attributes at the 0–20 cm depth: pH (in water) 6.1, 19.5 g kg<sup>-1</sup> organic matter, 5.0 mg dm<sup>-3</sup> P, 0.21 cmol<sub>c</sub> dm<sup>-3</sup> K, 5.08 cmol<sub>c</sub> dm<sup>-3</sup> Ca, 0.91 cmol<sub>c</sub> dm<sup>-3</sup> Mg, 2.6 cmol<sub>c</sub> dm<sup>-3</sup> H+Al, 0.0 cmol<sub>c</sub> dm<sup>-3</sup> Al, cation exchange capacity of 8.84 cmol<sub>c</sub> dm<sup>-3</sup>, and base saturation of 70%. A total of 2.0 kg dolomitic limestone, 5.0 kg simple superphosphate, and 0.5 kg potassium chloride were added to each cubic meter of soil (Espindula et al., 2015). To the other substrates, 6.0 g dm<sup>-3</sup> of the Basacot Plus 9M slow-release mineral fertilizer (Compo Expert Brasil Fertilizantes Ltda., Sumaré, SP, Brazil) were also added, containing 16% N, 8.0% P, 12% K, 2.0% Mg, 5.0% S, 0.4% Fe, 0.02% B, 0.02% Zn, 0.05% Cu, 0.06% Mn, and 0.015% Mo.

To obtain clonal seedlings of *C. canephora*, 5.0 cm long cuttings (orthotropic branch segments) of the BRS Ouro Preto cultivar were planted in the tubes with the substrates, which were continuously irrigated by automated nebulization at a 90–100% relative

humidity. At 35 and 70 days after staking (DAS), micronutrients were supplemented via fertigation by applying 0.125 g per plant of the Supra Mix Plus fertilizer (Supra Fertilizantes, Foz do Iguaçu, PR, Brazil), containing 5.0% Ca, 5.0% Zn, 4.0% Mn, 0.1% B, and 0.02% Mo, followed by the supplementation of 0.0125 g N per plant at 90 DAS.

At 120 DAS, the following parameters were evaluated: stem length, determined by bud insertion up to the apical meristem; stem diameter, obtained at the base of the branch, 2.0 cm above the insertion point of the shoot on the cuttings; root volume, considered the difference in displaced volume in a graduated beaker; stem dry mass (SDM), leaf dry mass (LDM), and root dry mass (RDM), determined on an analytical balance after drying in an oven with forced-air circulation, at 65°C; and total leaf area, using the free software to determine leaf area, Determinador Digital de Áreas (Ferreira et al., 2008). The following variables were, then, obtained: dry shoot mass (DSM = SDM + LDM), total dry mass (TDM = DSM + RDM), stem length/ diameter ratio (SLD = stem length/stem diameter), shoot/root ratio (SRR = DSM/RDM), and Dickson's quality index [DQI = TDM/(SLD + SRR)] (Dickson et al., 1960).

The Shapiro-Wilk test ( $p \le 0.05$ ) was used to evaluate data normality, followed by the analysis of variance. The means were grouped using the Scott-Knott test ( $p \le 0.05$ ), and, when significant differences were detected, the F-test ( $p \le 0.05$ ) was applied.

#### **Results and Discussion**

The physicochemical properties of the investigated substrates varied according to their composition, with greater differences being observed when each substrate was evaluated separately, i.e., from T1 to T5 (Table 2). The substrate formed only by coffee husk showed the highest values for electrical conductivity, dry density, buffering water, and remaining water, but the lowest for aeration space, available water, and RAW. Although presenting a lower pH, the sugarcane substrate stood out for its lower density and higher total porosity, available water, and RAW. The commercial substrate showed a high density and water holding capacity (WHC) according to the values obtained for available water, RAW, and buffering water. Vermiculite presented the lowest electrical conductivity, a low density, and the highest pH associated with a high

buffering water. Most of the combinations formed by these substrates showed similar values for the studied physicochemical variables. The SC+VM combination, for example, resulted in a lower total porosity, with a low aeration space and remaining water.

Although the evaluated physicochemical properties have been previously analyzed and their standards and ranges defined to characterize ideal substrate conditions for seedling production in containers (De Boodt & Verdonck, 1972; Kratz et al., 2013; Zorzeto et al., 2014; Schafer et al., 2015), studies are not yet conclusive concerning *C. canephora*.

In the case of total porosity, which is divided into aeration space (represented by macropores) and available water (divided into RAW and buffering water), the standard values are between 20 and 30% of the substrate volume for aeration space and 25 or 35% for available water, comprising 20 or 30% of RAW and 5.0% of buffering water (De Boodt & Verdonck, 1972). In the present work, wide variations in total porosity were observed in the treatments due to the different particle arrangements resulting from

the mixed materials. Sugarcane was important for the porosity of the substrate, increasing total porosity in some combinations up to values close to the 85% recommended for a substrate volume containing 15% solids (De Boodt & Verdonck, 1972).

Density and WHC are inversely proportional, as noted for the sugarcane substrate. Comparatively, very dense substrates present a lower total porosity and, therefore, retain little water, which impairs root development, whereas very low-density substrates do not promote an adequate plant fixation and balance in containers (Zorzeto et al., 2014). Kratz et al. (2013) pointed out that substrate density must be defined according to the height of the container, with recommended values ranging from 200 to 400 kg m<sup>-3</sup> for containers up to 15 cm high, as in the case of the tubes used in the present study.

For the pH of the substrate, the ideal values are from 5.0 to 6.5 according to Schafer et al. (2015). A pH value above the ideal was obtained for coffee husk and vermiculite, as well as for their combinations; however, the addition of these materials to the

**Table 2.** Electrical conductivity (EC), hydrogen potential (pH), dry density (DS), total porosity (TP), aeration space (AS), available water (AW), readily available water (RAW), buffering water (BW), and remaining water (RW) for each evaluated substrate used in the production of clonal *Coffea canephora* seedlings<sup>(1)</sup>.

Treatment <sup>(2)</sup>	EC	pН	DS	TP	AS	AW	RAW	$_{\mathrm{BW}}$	RW	
	(dS m <sup>-1</sup>	$H_2O$	(kg m <sup>-3</sup> )	(%)						
T1: CH	3.81a	6.59c	362.85a	77.89d	20.12e	3.18d	1.20d	1.97a	54.58a	
T2: EG	2.24d	5.64j	202.94e	77.13d	28.97c	13.66b	12.78b	0.88c	34.48f	
T3: SC	2.26d	5.491	140.23g	87.55a	31.10b	23.12a	21.77a	1.35b	33.32g	
T4: CS	0.43j	5.61j	365.48a	73.43e	18.91e	22.24a	19.74a	2.49a	32.27g	
T5: VM	0.041	6.90a	151.61g	84.80b	27.61c	11.91c	9.71c	2.20a	45.27b	
T6: CH+ EG	2.96c	6.34e	265.89b	78.58d	31.61b	9.90c	9.67c	0.23c	37.06e	
T7: CH+SC	3.24b	6.33e	245.93c	84.49b	30.26b	15.30b	14.88b	0.41c	38.92d	
T8: CH+CS	2.36d	6.43d	352.14a	74.08e	23.87d	14.73b	14.37b	0.35c	35.47f	
T9: CH+VM	1.66g	6.73b	220.84d	81.34c	31.25b	10.25c	9.77c	0.47c	39.83d	
T10: EG+SC	2.15e	5.73i	173.63f	86.03a	34.82a	13.95b	13.38b	0.57c	37.25e	
T11: EG+CS	1.52h	5.65j	261.15b	71.94f	26.17c	15.01b	14.44b	0.57c	30.76g	
T12: EG+VM	1.20i	5.91h	186.86f	74.89e	22.95d	12.71b	12.17b	0.53c	39.22d	
T13: SC+CS	1.15i	5.62j	217.70d	70.59f	24.36d	13.49b	11.32c	2.16a	32.72g	
T14: SC+VM	1.05i	5.96h	148.89g	62.44g	18.55e	10.56c	9.39c	1.16c	33.32g	
T15: CH+SC+EG	2.75c	6.16f	230.97d	76.76d	31.60b	3.14d	1.28d	1.85a	42.02c	
T16: CH+SC+EG+CS	2.06e	6.05g	243.26c	76.23d	33.82a	8.71c	8.52c	0.18c	33.69g	
T17: CH+SC+EG+CS+VM	1.80g	6.08g	227.37d	73.82e	27.36c	13.85b	13.29b	0.55c	32.61g	
T18: CH+SC+VM	1.58h	6.35e	181.51f	75.59d	32.58b	9.53c	9.46c	0.06c	33.47g	
T19: CH+EG+VM	2.08e	6.29e	251.41c	74.02e	28.32c	10.01c	9.49c	0.52c	35.69f	
T20: CH+EG+SC+VM	1.93f	6.17f	192.30e	73.42e	28.78c	10.33c	9.87c	0.45c	34.31f	
Coefficient of variation (CV, %)	4.06	0.56	3.59	1.57	5.14	10.54	11.65	34.57	3.65	

<sup>(1)</sup>Means followed by equal letters do not differ by the Scott-Knott test, at 5% probability. (2)CH, coffee husk; EG, elephant grass; SC, sugarcane; CS, commercial substrate; and VM, vermiculite.

commercial substrate, sugarcane, or elephant grass may be a strategy to adapt substrate pH.

Although the analyzed substrates influenced coffee seedling growth, they did not differ significantly from each other. The substrates composed of just one material (T1, T2, T3, T4, and T5) did not promote seedling growth (Table 3). Moreover, the commercial substrate (T4) and the standard (T21) showed similar values for most of the evaluated parameters, except for stem length, which was lower in the latter.

The physicochemical characteristics of the coffee husk substrate affected seedling growth, resulting in the lowest stem length, leaf area, and SLD, all with values lower than those obtained for the standard (Table 3). Therefore, coffee husk is viable for use as a substrate only when mixed with other materials, especially vermiculite and except elephant grass. Furthermore, the high values found for electrical conductivity, density, and remaining water and the low ones for aeration space, available water, and RAW

make that residue unfavorable for coffee seedling growth.

Regarding remaining water, the value obtained for the coffee husk substrate differed from that of the other materials (Table 2). This variable, equivalent to the water remaining in a substrate after the application of a 100 kPa tension, is considered ideal between 20 and 30% of the substrate volume (De Boodt & Verdonck, 1972; Zorzeto et al., 2014). According to Kratz et al. (2013), values above 30% lead to a poor water drainage, which can damage root growth. The coffee husk material also presented a high electrical conductivity, which may be associated with its high salt content, especially of nitrogen and potassium (Table 1). The combination of the high values of remaining water and of electrical conductivity may have hindered salt percolation, culminating in an excessive salinity in the solution (Schafer et al., 2015). Temoteo et al. (2015) concluded that the initial growth of C. canephora seedlings is impaired from 2.0 dS m<sup>-1</sup> water salinity. When coffee husk was mixed with the other substrates,

**Table 3.** Stem length (SL), stem diameter (SD), root volume (RV), leaf area (LA), and stem length/diameter ratio (SLD) of *Coffea canephora* seedlings produced in different substrates<sup>(1)</sup>.

Treatment <sup>(2)</sup>	SL	SD	RV	LA	SLD
	(cm)	(mm)	(cm <sup>3</sup> )	(cm <sup>2</sup> )	-
T1: CH	6.95d	3.28b	0.51b	81.62c	2.09c
T2: EG	16.67b	4.01a	2.08b	337.82a	4.17b
Г3: SC	15.22c	3.67b	1.35b	258.72b	4.09b
Г4: CS	16.15b	3.73b	1.51b	276.20b	4.30b
Γ5: VM	16.79b	3.88b	1.93b	303.70b	4.30b
Γ6: CH+ EG	16.93b	3.77b	1.59b	267.09b	4.52a
Γ7: CH+SC	18.17b	4.17a	2.30a	400.59a	4.38b
T8: CH+CS	18.05b	4.01a	2.23a	370.27a	4.51a
Г9: СН+VМ	18.60b	4.23a	2.53a	386.96a	4.42b
Γ10: EG+SC	17.37b	3.85b	1.68b	350.69a	4.51a
Γ11: EG+CS	19.91a	4.31a	2.83a	466.86a	4.60a
Γ12: EG+VM	21.60a	4.31a	2.90a	425.02a	5.02a
Γ13: SC+CS	19.91a	4.13a	2.44a	385.88a	4.88a
Γ14: SC+VM	21.25a	4.31a	3.25a	429.17a	4.95a
T15: CH+SC+EG	19.64a	3.88b	1.99b	360.07a	5.03a
Γ16: CH+SC+EG+CS	15.02c	3.80b	1.76b	272.81b	3.97b
Γ17: CH+SC+EG+CS+VM	17.86b	4.32a	2.33a	402.26a	4.08b
Г18: CH+SC+VM	23.04a	4.43a	3.17a	503.54a	5.21a
Γ19: CH+EG+VM	14.90c	4.08a	2.74a	315.88b	3.69b
Γ20: CH+EG+SC+VM	19.63a	4.20a	2.72a	426.98a	4.67a
Γ21: soil	13.25c	3.62b	1.24b	252.79b	3.73b
Coefficient of variation (CV, %)	15.87	8.36	38.91	26.17	13.57

<sup>(1)</sup>Means followed by equal letters do not differ by the Scott-Knott test, at 5% probability. (2)CH, coffee husk; EG, elephant grass; SC, sugarcane; CS, commercial substrate; and VM, vermiculite.

the electrical conductivity values were also high, but not enough to significantly limit the growth of the seedlings, suggesting their tolerance to salinity levels of up to 2.3 dS m<sup>-1</sup>. The mixture of coffee husk with vermiculite, however, reduced the conductivity values of the resulting substrate.

The dry masses (SDM, RDM, LDM, DSM, and TDM) were higher for the combined substrates, except in treatments T6, T10, T15, T16, and T19, when compared with the substrates evaluated separately (T1–T5) and the standard (T21) (Table 4). Regarding the SRR, the highest average was observed for the coffee husk substrate and the lowest, for the commercial substrate, vermiculite, and all substrate combinations except CH+EG and EG+SC. For this variable, the elephant grass and sugarcane treatments did not differ from the standard.

As to the studied ratios and index, Marana et al. (2008) reported values from 3.5 to 4.0 for the SLD, 4.0 to 7.0 for the SRR, and higher than 0.2 for the DQI when evaluating seminiferous *Coffea arabica* L.

seedlings grown in 120 cm<sup>-3</sup> tubes. These values have been previously used for comparisons with those of C. canephora indices (Dardengo et al., 2013; Pereira et al., 2013), as there are no known specific standards in the literature for seedlings of this species originated from cuttings and produced in 280 cm<sup>-3</sup> plastic tubes. In the present work, the SLD values, except for coffee husk, were higher than those found by Marana et al. (2008), which may suggest the possibility of seedling etiolation. However, SDM and LDM values higher than or similar to those of the control indicate a satisfactory and proportional stem development in relation to the other aerial plant organs. Therefore, it is suggested that the SLD reference values for C. canephora clonal seedlings produced in 280 cm<sup>-3</sup> tubes may range from 4.5 to 5.2.

In the coffee husk substrate, the SLD was the lowest, even in comparison with that of the standard, due to the shorter stem length observed in that treatment. Therefore, when there is an adequate relationship between stem length and diameter, the plants are

**Table 4.** Stem dry mass (SDM), root dry mass (RDM), leaf dry mass (LDM), shoot dry mass (DSM), total dry mass (TDM), shoot/root ratio (SRR), and Dickson's quality index (DQI) obtained for *Coffea canephora* seedlings produced in different substrates<sup>(1)</sup>.

Treatment <sup>(2)</sup>	SDM	RDM	LDM	DSM	TDM	SRR	DQI
			(g)				
T1: CH	0.17b	0.09b	0.37b	0.52b	0.62b	5.40a	0.08c
T2: EG	0.61b	0.59b	1.53b	2.05b	2.74b	3.79b	0.34b
T3: SC	0.53b	0.48b	1.20b	1.73b	2.21b	3.72b	0.27b
Γ4: CS	0.51b	0.60b	1.25b	1.70b	2.30b	2.98c	0.31b
Γ5: VM	0.54b	0.69b	1.43b	1.98b	2.67b	3.01c	0.36b
Γ6: CH+ EG	0.55b	0.46b	1.18b	1.63b	2.20b	3.95b	0.26b
Γ7: CH+SC	0.78a	0.85a	1.80a	2.58a	3.44a	3.41c	0.47a
Γ8: CH+CS	0.69a	0.69b	1.75a	2.45a	3.15a	3.59c	0.39b
Г9: CH+VM	0.75a	0.78a	1.88a	2.63a	3.41a	3.42c	0.45a
Γ10: EG+SC	0.65b	0.58b	1.57b	2.23b	2.81b	3.81b	0.33b
Γ11: EG+CS	0.86a	0.92a	2.12a	2.99a	3.91a	3.26c	0.50a
Γ12: EG+VM	1.06a	1.12a	2.24a	3.31a	4.43a	3.08c	0.56a
Γ13: SC+CS	0.74a	0.86a	1.85a	2.60a	3.47a	3.03c	0.44a
Γ14: SC+VM	0.92a	1.22a	2.26a	3.19a	4.42a	2.64c	0.61a
Γ15: CH+SC+EG	0.62b	0.68b	1.54b	2.17b	2.85b	3.18c	0.35b
Γ16: CH+SC+EG+CS	0.58b	0.53b	1.22b	1.74b	2.27b	3.30c	0.31b
Γ17: CH+SC+EG+CS+VM	0.71a	0.98a	1.91a	2.62a	3.61a	3.01c	0.51a
Γ18: CH+SC+VM	1.08a	1.15a	2.50a	3.59a	4.74a	3.33c	0.57a
Γ19: CH+EG+VM	0.55b	0.76a	1.47b	2.03b	2.79b	3.20c	0.43a
Г20: CH+EG+SC+VM	0.79a	1.03a	2.13a	2.85a	3.88a	2.82c	0.52a
Γ21: soil	0.41b	0.39b	1.11b	1.52b	1.91b	4.27b	0.25b
Coefficient of variation (CV, %)	33.12	44.45	31.54	31.81	34.18	19.82	41.51

<sup>(1)</sup>Means followed by equal letters do not differ by the Scott-Knott test, at 5% probability. (2)CH, coffee husk; EG, elephant grass; SC, sugarcane; CS, commercial substrate; and VM, vermiculite.

taller, but not because of seedling etiolation, and tend to present thicker stems. In addition, the higher values for the SRR in this substrate indicate an incipient root system development in relation to the shoot, resulting from the undesirable characteristics of coffee husk discussed previously.

The SRR values obtained in all treatments, except in T1, are below those reported by Marana et al. (2008) for fertilized coffee plants, which would indicate a deficient aerial seedling formation in relation to the roots. However, based on the adequate stem length and DSM values observed in most treatments, it can be inferred that the SRR values are due to the larger tube volume, providing a better root system development and implying in a greater similarity between DSM and RDM values. Silva et al. (2013) highlighted that a greater substrate space and volume provide an adequate root system development by allowing of a better root access to moisture and nutrients, minimizing stress due to the lack of water and enabling the maintenance of the aerial plant part.

The DQI was significantly higher in the substrate combinations, except in treatments T6, T8, T10, T15, and T16 (Table 4). In T1, the unfavorable physicochemical characteristics of the coffee husk substrate led to a lower seedling DQI. The DQI, as well as the SRR, are good seedling quality indicators as phytomass distribution balance (SLD and SRR) and robustness (TDM) are considered for both respective calculations (Dardengo et al., 2013). Therefore, the DQI values above 0.2 in all treatments, except in the one with coffee husk, reinforce that the evaluated substrates promote an adequate *C. canephora* seedling development.

Seedling growth in the substrate composed only of soil did not differ from that in the commercial substrate. In addition, the values obtained in all treatments, except in T1, were superior to those found by Berilli et al. (2014) for seedlings of the same age grown in a substrate mixed with the soil matrix, indicating adequate conditions for seedling development.

The substrates in treatments T7, T9, T11, T12, T13, T14, T17, T18, and T20 overlapped with the others for all tested variables, not differing from each other, being, therefore, indicated for the production of clonal *C. canephora* seedlings. Although coffee husk presented unsatisfactory results when used alone, it was also promising when combined with vermiculite, meaning

it should not be excluded from substrate mixtures for the production of clonal *C. canephora* seedlings.

#### **Conclusions**

- 1. Combining coffee husk, sugarcane, and elephant grass with each other and with vermiculite and commercial substrate improves the quality of clonal *Coffea canephora* seedlings.
- 2. Coffee husk, when combined with vermiculite up to a proportion of 50%, is suitable for clonal *Coffea canephora* seedling production.
- 3. Adding coffee husk, sugarcane, and elephant grass to standard substrate (subsurface soil) improves the quality of clonal *C. canephora* seedlings.

### Acknowledgments

To Consórcio Brasileiro de Pesquisa e Desenvolvimento do Café (CBP&D/Café), for financial support (project number 02.13.02.036.00.00) and to Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq), for research productivity grant of the corresponding author.

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