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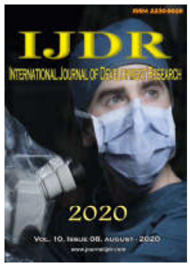

RESEARCH ARTICLE

OPEN ACCESS

MORPHOLOGICAL INDICES OF OKRA SEEDLINGS PRODUCED WITH COFFEE GROUNDS AND EGGSHELLS

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ABSTRACT

The okra has been highlighted for its nutritional potential and easy cultivation in small properties and in the so-called urban agriculture. Residues such as coffee grounds and eggshell have proved to be excellent alternatives for organic fertilization. Therefore, the objective of this work was to evaluate the agronomic potential of coffee grounds with eggshell in the production of Santa Cruz okra seedlings 47. The work was developed in the city of Chapadinha – MA, in a greenhouse, using a delineation completely randomized in a 4x3 factorial scheme for 21 days. Twelve treatments were used with 3 repetitions. The variables that gave significance between the factors, were explored by the Scott Knott test. Significance assessments described that there was no significant effect of the interaction between factors for any of the variables analyzed. However, there was a significant effect, alone, of the coffee grounds factor on the leaf area and the leaf area ratio. The higher the concentrations of coffee grounds, the lower results were obtained for the respective variables. Eggshell did not influence okra seedlings. It is necessary to carry out more research on the use of these residues in the production of vegetable seedlings.

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INTRODUCTION

Okra (*Abelmoschusesculentus* (L.) Moench) is a plant of African origin belonging to the *Malvaceae* family, it is being widely cultivated in tropical regions of the world due to favorable climatic conditions for its cultivation (Gemede *et al*., 2015, Santos *et al*., 2018a). The okra fruit has a great nutritional value, as it is rich in proteins, potassium, calcium, iron, phosphorus, vitamins A and B, in addition to 30% of the recommended levels of vitamin C (Gemede *et al*., 2015, Adekiya *et al*., 2019). In this way, their production becomes important to guarantee food security in various places in the world (Adekiya *et al*., 2019). Seedling production is one of the most important stages in the production of many agricultural systems, and in recent years we have chosen to use appropriate containers and alternative quality substrates to replace land use, which has enabled substantial increases in seedling quality. (Goncalves *et al*., 2018). A substrate to be considered of quality must guarantee a high germination rate, excellent plant growth, supply nutrients and have a good moisture retention capacity (Soares *et al*., 2020). Currently, there is a great interest in the agricultural environment for the use of organic residues in the production of seedlings, since these residues can supply nutrients and organic matter to plants, which promotes the development of an increasingly sustainable agriculture (Soares *et al*., 2020). Even though there are commercial substrates of excellent quality for the production of seedlings of certain species, there is a constant demand for new sustainable production technologies that meet the needs of crops and at the same time the economic conditions of producers are accessible (Ferreira *et al*., 2018). In this perspective, the use of ecological-based waste in the formulation of substrates appears as an alternative source for agricultural production (Zeist *et al*., 2019). Brazil ranks second in the world among the largest coffee consumers, with 21 million bags per year, behind only the United States (ABIC, 2019). Each ton of soluble coffee produced generates around 450 kg of sludge (Martinez-Saez *et al*., 2017), making it a possible environmental pollutant. With this in mind, attempts to recycle sludge are being tested in the composition of substrates for the production of seedlings, as it contains high concentrations of organic and inorganic compounds, giving it great agricultural potential (Garcia *et al*., 2016). Coffee grounds contain essential nutrients in plants, such as: organic matter (90.46%), nitrogen (2.30%), phosphorus (0.15%), potassium (0.35%) and magnesium (0.13%) (Martinez-Saez *et al*., 2017). Regarding eggshells, 44.2 billion units of eggs were produced in Brazil in 2018 (ABPA, 2019). About 99% of this production supplies the domestic market, generating large discards of egg shells in domestic waste. In this sense, there is a concern about the incorrect disposal of eggshell since it contains a rich membrane that favors microbiological activity, which can affect public health (Vieira *et al*., 2017). Ferreira *et al*. (2020) point out that around 6 million tons of this waste are discarded worldwide annually. According to Rodrigues and Ávila (2017), the composition of the eggshell is 94% calcium carbonate, 1% calcium phosphate, 4% organic compounds and 1% magnesium carbonate, thus presenting potential as a fertilizer. In addition, the use of eggshell in the agricultural scenario is mainly aimed at pH correction, given its high content of calcium carbonate ($CaCO_3$) present in its composition (Lo Mônaco *et al*., 2015). Thus, in order to contribute to the development of okra cultivation in the State of Maranhão, the objective of this study was to evaluate the effect of the joint use of coffee grounds and eggshell on the production of the Santa Cruz 47 okra seedlings.

MATERIALS AND METHODS

The experiment was installed and conducted in a greenhouse with 50% luminosity control, from 05 to 26 September 2019, at the Center for Agricultural and Environmental Sciences of the Federal University of Maranhão, in Chapadinha-MA (03º44'17 "S and 43º20'29 "W and altitude of 107 m). The soil used was classified as dystrophic Yellow Latosol (LAd) (Santos *et al*., 2018b). The region's climate is classified by Köppen as Aw, humid tropical (Selbach & Leite, 2008), with total annual precipitation ranging between 1,600 to 2,000 mm (Nogueira *et al*., 2012) and average annual temperature above 27 ° C (Passos *et al*., 2016). A completely randomized design with a 4x3 factorial scheme (four proportions of coffee grounds and 3 eggshell proportions) was used, totaling 12 treatments, where each treatment had the following formulations: T1: 5% of coffee grounds (CG) + 5% eggshell (E) + 90% dystrophic Yellow Latosol (LAd), T2: 5% CG + 10% E + 85% LAd, T3: 5% CG + 15% E + 80% LAd, T4: 10% CG + 5% E + 85% LAd, T5: 10% CG + 10% E + 80% LAd, T6: 10% CG + 15% E + 75% LAd, T7: 15% CG + 5% E + 80% LAd, T8: 15% CG + 10% E + 75% LAd, T9: 15% CG + 15% E + 70% LAd, T10: 20% CG + 5% E + 75% LAd, T11: 20% CG + 10% E + 70% LAd, T12: 20% CG + 15% E + 65% LAd. Each treatment had 3 replicates of 9 seedlings each, totaling 324 experimental plots. The physical and chemical analyzes of the LAd were carried out at the Soil Science Laboratory of the Federal University of Ceará, Fortaleza-CE. In the chemical analysis (Table 1), pH, electrical conductivity (EC) and the total contents of the macronutrients were analyzed: nitrogen (N), phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg) and sulfur (S) according to MAPA (2007). Through the granulometryanalysis it was found that the LAd has: 384 g coarse sand kg^{-1}, 336 g fine sand kg^{-1}, 112 g of silt kg^{-1}, 168 g of total clay kg^{-1}, 38 g of natural clay kg^{-1}, Franco sandy texture, and flocculation degree of 0.77 kg kg^{-1}. In the physical analysis (Table 2), the following were analyzed: global density (GD), particle density (PD) and porosity (%), determined according to the procedures described by Schmitz *et al*. (2002). Prior to the formulation of the substrates, the LAd was sieved in a 8 mm mesh sieve to remove stony, branches and leaves, to facilitate its homogenization. In addition, the LAdwas sterilized in an autoclave. The eggshell (E) was ground in a manual pestle and then crushed in a blender to reduce the granulomentry and facilitate incorporation into the substrate. Coffee grounds (CG) were obtained in several coffee consuming establishments in the state of Maranhão, without distinction of brand or specific type, and placed to dry in the shade until the moisture present was eliminated. Subsequently, both CG and E were mixed with LAd according to the proportions of each treatment. The formulated substrates were placed in two polyethylene germ trays of 162 cells each and a volume of 50 cm^3 per cell, sowing three seeds per cell.

At 7 days after sowing (DAS), thinning was carried out, leaving only the most vigorous plant in each cell. Irrigation was carried out twice a day, early in the morning and late afternoon, using a 20 ml $seedling^{-1}$ daily slide. At 21 DAS, the following were evaluated: a) leaf area (LA): measured in cm2 and obtained using the ImageJ® software, b) leaf area ratio (LAR): obtained by the ratio between the leaf area and the total dry mass of the plant, c) root thickness (RT): obtained by the ratio between root dry mass and root length, d) Root specific length (RSL): obtained by the ratio of root length and root dry mass, and e) plant height / shoot dry mass ratio (PH/SDMR): obtained by the ratio of plant height to shoot dry mass. The data obtained were subjected to analysis of variance for diagnosis of significant effect ($P \leq 0.05$) using the computer program Infostat® version 20151 (DiRienzo*et al*., 2015). With significance between the factors, the data were explored using the Scott Knott test ($\alpha = 0.05$). With significance for the factors individually, polynomial regression analysis was performed, the equations being selected by the F test ($P < 0.05$).

RESULTS AND DISCUSSION

The analysis of variance did not find a significant effect ($P > 0.05$) of the CGxE interaction for any observed variable. Likewise, there was no significance ($P > 0.05$) of the eggshell factor for any variable. However, there was a significant effect ($P < 0.05$) of the coffee grounds factor for the leaf area and the leaf area ratio (Table 3). Similar results were found by Ferreira *et al.* (2020), who found no significant effect for CGxE for numerous parameters of okra seedlings. The LA variable adjusted to the decreasing linear model, that is, it was found that the increasing concentrations of coffee grounds inhibited the development of the leaf area of the okra, with a linear decrease as the concentration of CG increases in the substrate formulation (Figure 1). Kitou and Yoshida (1997) stated that it is not appropriate to directly apply coffee residues as fertilizers, as they present harmful compounds to vegetables.

Table 1. Values of pH, electrical conductivity (EC) and total contents of nitrogen (N), phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg) and sulfur (S) of the dystrophic Yellow Latosol

Substrate	pH	CE	N	P	K	Ca	Mg	S
		dS m^{-1}	g Kg^{-1}	mg Kg^{-1}		cmol$_c$ Kg^{-1}		
Lad	5.06	0.10	0.63	13	0.07	0.80	0.30	1.5

LAd= dystrophic Yellow Latosol.

Table 2. Global density (DG), particle density (DP) and porosity of the dystrophic Yellow Latosol

Substrate	GD	PD	Porosity (%)
	g cm^{-3}		
LAd	1.44	2.67	45.99

LAd= dystrophic Yellow Latosol.

Table 3. Summary of analysis of variance of the variables leaf area (LA), leaf area ratio (LAR), root thickness (RT), specific root length (SRL) and plant height / shoot dry mass ratio (PH/SDMR) of okra seedlings cv. Santa Cruz 47 submitted to different concentrations of coffee grounds and eggshell in the soil

VS	DF	Medium Square				
		LA	LAR	RT	SRL	PH/SDMR
CG	3	17314.76 *	7795.15 *	4.2^{-04ns}	2877.99 ^{ns}	228.77 ^{ns}
E	2	4171.35 ^{ns}	209.92 ^{ns}	4.5^{-04ns}	4983.30 ^{ns}	56.97 ^{ns}
CG x E	6	3836.28 ^{ns}	1618.39 ^{ns}	1.8^{-04ns}	2726.65 ^{ns}	267.28 ^{ns}
Residue	23	1857.71	1035.16	2.1^{-04}	2375.34	183.05
CV (%)		44.14	18.93	56.36	79.08	49.37

VS: Variation source. DF: Degree of freedom. CG: Coffee grounds. E: Eggshell. CV: Coefficient of variation. *: significant at 5% probability.^{ns}: not significant by the test Scott Knot.

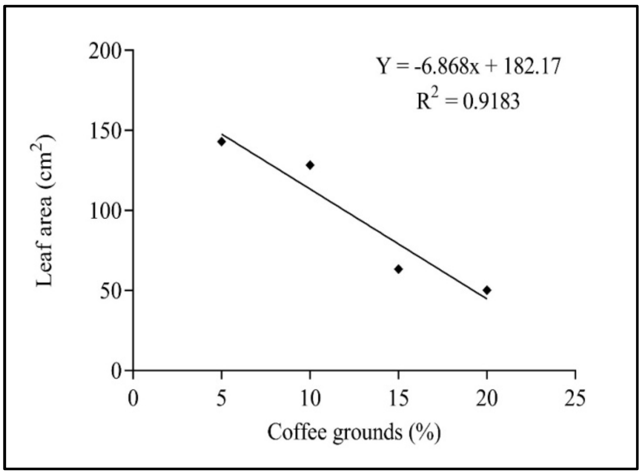

Figure 1. Leaf area (cm^2) of okra seedlings cv. Santa Cruz 47 submitted to different concentrations of coffee grounds and eggshell in the soil

The same authors report that it is necessary to process the CG, either by composting or vermicomposting, to use this residue in the agricultural environment. Others point to the use of fresh and treated coffee grounds to improve the antioxidant capacity of lettuce (Cruz *et al*., 2014). Tuntiwiwattanapun and Tongcumpou (2018) and Kooks (2018) recommended the use of CG in agriculture since its composition contains elements with potential fertilizer. However, Carmo *et al.* (2018) highlight the need for CG composting before applying it in the formulation of substrates. Another viable alternative is the use of CG in the pyrolysis process forming the product known as biochar (Liu *et al*., 2017). Martins Filho (2017) reports an improvement in chemical attributes and an increase in pH with the use of CG biochar in neossol grown with corn and beans. LA is an excellent indicator of the productivity of a crop, since it is in the leaves that a good part of the photosynthetic process occurs, which involves a series of complex processes and reactions that occur during the growth and development of the plant (Taiz *et al*., 2017), so that the leaf surface is the basis of the potential yield of the crop. The light interception made by the canopy has a great influence on the production of the cultures, because it is through this leaf interception that the plant will be able to absorb the necessary amounts of incident energy, through the leaf interception (leaf area), which will later be absorbed (electronic excitation), converted (CO_2 fixation), be redistributed among the parts of the plant (translocation of photoassimilates) and metabolized in the different parts of the plant (efficiency in use) (Silva *et al*., 2011). In addition, knowing the PA of the plant is essential for agronomic and physiological studies, involving analysis of plant growth and transpiration (Ferreira *et al*., 2015). Assessing the leaf area allows a great help in the selection of genotypes in breeding programs, considering that, in addition to direct selection based on productivity, other strategies can be used in order to maximize the gains with the selection, reducing the demand for time and resources (Cruz *et al*., 2004). Therefore, the result obtained with the use of CG causes concern, because instead of promoting the development of LA, CG reduces the development of this variable. Analyzing the leaf area ratio (LAR) (*$P<0.05$*), it was found that it adjusted to the decreasing quadratic model, the highest value being obtained with the concentration of 15% CG in the substrate (Figure 2).

The LAR allows us to estimate the useful leaf area, that is, the photosynthetically active area, so that with the growth of the plant there is a decrease in values, due to the increase in shading, which affects the useful leaf area for the photosynthesis (Benicasa, 2003). It was observed that CG promotes the growth of LAR until the concentration of 15% of CG present in the substrate formulation. Values above 15% of CG in the substrate induce a decrease in LAR due to a possible phytotoxicity of CG.

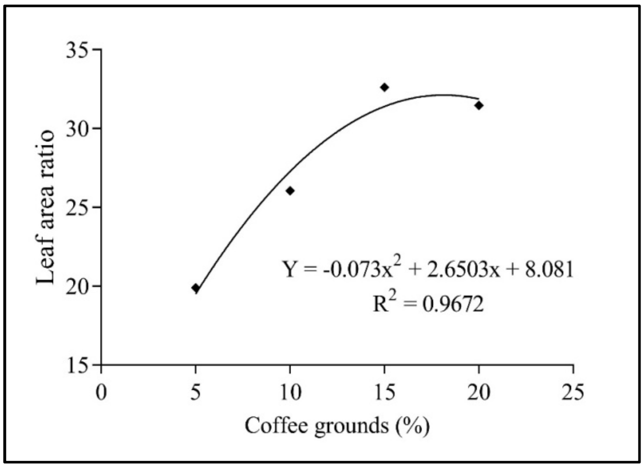

Figure 2. Leaf area ratio (cm^2 g^{-1}) of okra seedlings cv. Santa Cruz 47 submitted to different concentrations of coffee grounds and eggshell in the soil

The results obtained are similar to those found by Cruz (2015), who found a decrease in the growth and development of lettuce, carrots and spinach. According to the author, this inhibition possibly occurred due to the presence of caffeine, which may have reduced the amount of nitrogen absorbed by the plants or even inhibited the development of these vegetables. Despite the results obtained, the results were not homogeneous, with the RAF growing with the use of the tested BC concentrations, while the AF showed inhibition with the same doses. The other variables did not respond to CGxE interaction. Therefore, it is clear that there is a need to conduct more research for the use of these residues in the production of seedlings, since they have great potential for polluting the environment.

Conclusion

The interaction of coffee grounds with eggshell does not influence the morphological indexes of Santa Cruz okra seedlings 47. Coffee grounds promote the inhibition of leaf area development and the leaf area ratio of Santa Cruz okra seedlings 47. It is necessary to carry out further research on the use of these residues in the production of seedlings, in order to develop new concentrations, methodologies of use and purposes for these residues, aiming to develop an increasingly sustainable agriculture.

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