








Increasing levels of cassava wastewater in the production of papaya formosa seedlings

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Abstract

Cassava wastewater (CWW) is rich in nutrients for plants, presenting great potential as an alternative fertilizer. However, this waste is incorrectly disposed of in the environment, which causes environmental problems. The objective was to evaluate the influence of increasing doses of CWW on the production of formosa papaya seedlings. The experiment was installed in a greenhouse from October 22 to December 5, 2019, in which a completely randomized design was applied with 4 treatments of different levels of CWW with 0 ml L⁻¹, 6.25 ml L⁻¹, 12.5 ml L⁻¹, 25 ml L⁻¹, where each treatment contained 4 replicates of 4 seedlings. Biometric parameters were evaluated to identify the effects of CWW on initial papaya growth. Data were subjected to analysis of variance by F test, and data were explored through regression analysis. Significance was found for root length (RL) and root diameter (RD). The other biometric parameters did not respond to the tested doses of CWW. The tested doses reduced the RL and the DR had the best performance with the dose of 13.5 ml L⁻¹. 13.5 ml L⁻¹ of cassava effluents is recommended for better development of DR. In general, this residue has an inhibitory effect on the initial growth of the papaya tree and is not recommended for the production of seedlings of this species.

Keywords: *Carica papaya* L., plant nutrition, sustainable agriculture, use of residues

Introduction

Papaya is widely consumed in the world due to its flavor, nutritional value and various uses in the food industry. This significantly leveraged its production worldwide (Chan-Léon et al., 2017), as it is well adapted to tropical and subtropical climates, with great expression in the economy of the Brazilian Northeast (Sá et al., 2013).

According to data from the Food and Agriculture Organization (FAO, 2021), in 2020, Brazil revealed that it has great potential for the cultivation of papaya, as it produced about 1.2 million tons of papaya that year, which put it among the largest producers of the fruit in the world.

A viable alternative to boost fruit production would be to promote the development of this crop through technologies adapted to each region (Gruda et al., 2019; Gruda, 2019), since Brazil has continental proportions that guarantee a heterogeneity of conditions

edaphoclimatic.

Papaya cultivation requires constant replacement of the orchards (from two and a half to four years), which promotes a great demand for papaya seedlings (Faria et al., 2009). The production of seedlings is one of the most important stages of the fruit system, as it directly influences the initial formation of orchards (Guedes et al., 2010) and, therefore, the final production.

Residues from agro-industrial productions can pollute soils and water bodies, causing serious problems in several ecosystems, affecting not only the environment, but also human health (Nóbile et al., 2017). With that in mind, the reuse of these wastes becomes crucial. World agriculture and companies no longer consider waste as disposable materials, but as raw materials for the production chain and alternative procedures. Thus, it is important to find approaches that promote the use of organic waste in production chains (Chrysargyris et al.,

2021).

Among these residues is cassava wastewater (CWW), an effluent from the processing of cassava roots with a pale-yellow color and milky consistency, containing several substances that bring benefits to vegetables (Chitwood et al., 2005; Ferreira et al., 2015). As it is among the largest producers of cassava in the world (FAO, 2021), it is assumed that there is a large production of this residue in the country and, consequently, its incorrect disposal in the environment.

Numerous studies highlight the agricultural potential of this effluent, which has potential as a fertilizer (Barreto et al., 2014; Duarte et al., 2014) and in the management of insect pests (Natsu et al., 2016; Pinto-Zevallos et al. 2018). Thus, it is opportune to evaluate the potential of this effluent as a stimulant in the initial growth of commercial species, such as papaya.

Thinking about it and with the objective of contributing to the reuse of agricultural waste, the objective of this work was to evaluate the effect of different concentrations of CWW on the production of papaya seedlings Formosa.

Material and Methods

The experiment was conducted from October 22 to December 5, 2019, in a greenhouse located in a flat relief with 50% light control, installed at the Chapadinha Science Center of the Federal University of Maranhão, located in the municipality of Chapadinha-MA (03°44'17"S and 43°20'29"W and altitude of 107 m). The climate of the region is classified as humid tropical (Aw) according to the Köppen classification, with an average annual rainfall of 1,613.2 mm and an average annual temperature of 27.9°C (Passos et al., 2016). Weather conditions during the experimental period can be seen in (Figure 1).

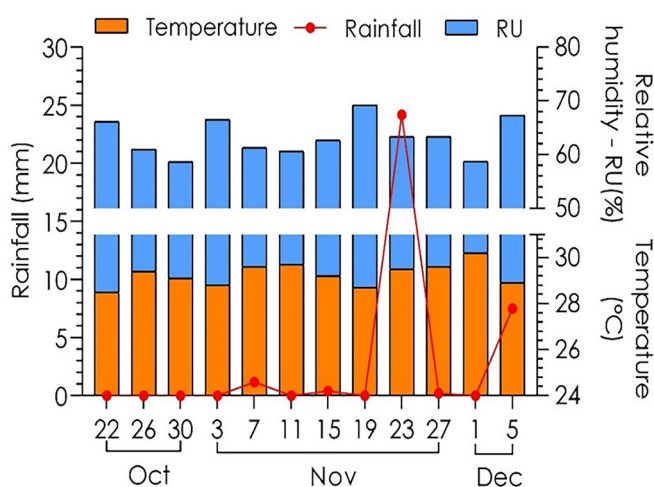


Figure 1. Climatic conditions of the experimental area during the condition period of the experiment.

A completely randomized design with 4 treatments was adopted, where each treatment corresponded to the following levels of cassava wastewater: T1: 0 ml of cassava wastewater L⁻¹ of water; T2: 6.25 ml L⁻¹; T3: 12.5 ml L⁻¹; T4: 25 ml L⁻¹. Four applications were carried out with the aid of a manual sprayer with a capacity of 1 liter at an interval of 5 days between each application, with the first application being made at 28 days after sowing (DAS). Each treatment contained 4 replicates, with each replicate having 4 seedlings.

The substrates used were composed of 40% carbonized rice straw (CRS) + 40% soil + 20% decomposed babassu stem (DBS) (Cordeiro, et al. 2020). The soil used was classified as Districohesive Yellow Latosol (LAdx) (Santos et al., 2018).

The physical (global density, particle density and porosity) and chemical (pH, electrical conductivity and total contents of nitrogen, phosphorus, potassium, calcium, magnesium and sulfur) of the materials used are presented in (Table 1).

Cassava was obtained from Associação dos Agricultores de São Gonçalo in the municipality of Santana do Maranhão (3° 06' 57" S and 42° 24' 43" W and altitude of 43 m), during the processing stage of cassava roots to flour production. The cassava wastewater was stored in 500 liter polyethylene reactors for 60 days at room temperature. Chemical characterization was performed according to the procedures of the American Public Health Association (APHA, 1995), obtaining: pH = 4.8; organic matter = 2.7%; N = 330.0 mg L⁻¹; P = 240.0 mg L⁻¹; K = 3450 mg L⁻¹; Ca = 228.0 mg L⁻¹; Mg = 1000.0 mg L⁻¹; S = 42.0 mg L⁻¹; Cu = 0.430 mg L⁻¹; Fe = 1740 mg L⁻¹; Mn = 0.4 mg L⁻¹; Zn = 0.39 mg L⁻¹ and Na = 40 mg L⁻¹.

Prior to the formulation of the substrates, the rice straw was carbonized and the decomposed babassu stem was sieved in a sieve with a 8 mm mesh, to facilitate its homogenization, and later they were mixed with the soil according to the proportions of 40% of CRS + 20% DBS + 40% LAdx and placed in polyethylene bags measuring 12 x 20 cm, sowing two seeds per bag.

Irrigation was performed twice a day with the aid of a manual watering can with a capacity of 5 L. At 28 DAS, thinning was performed, leaving only the most vigorous plant in each container.

The experiment was evaluated at 45 DAS, where the primary variables were determined: i) plant height (PH, cm): determined from the substrate level to the apex of the seedling with the aid of a millimeter ruler; ii) stem diameter (SD, mm): obtained with a digital caliper (Digimess®) at the substrate level; iii) number of sheets (NF):

Table 1. Chemical and physical characterization of the dystrocohesive Yellow Latosol (LAdx), carbonized rice straw and the decomposed babassu stem (DBS)

Sub.	pH	EC dS m ⁻¹	N – mg kg ⁻¹ –	P	K	Ca — cmol _c Kg ⁻¹ —	Mg	S	GD – g cm ⁻³ –	PD	PR %
LAdx	5.06	0.10	630	13	0.07	0.80	0.30	1.5	1.44	2.67	45.99
CRS	7.90	6.13	7.00	3.06	15.97	7.40	18.20	42.3	0.39	1.29	69.70
DBS	5.32	4.34	5880	33	3.63	20.60	15.20	41.5	0.33	0.97	65.95

Sub.: substrates. pH: hydrogen potential. EC: electrical conductivity. N: nitrogen. P: phosphorus. K: potassium. Ca: calcium. Mg: magnesium. S: sulfur. GD: global density. PD: particle density. PR: porosity.

obtained by manually counting sheets; iv) root length (RL, cm): measured with a graduated ruler; v) root volume (RV, cm³): performed by measuring the displacement of the water column in a graduated cylinder; vi) leaf area (cm²): quantified using the imageJ[®] computer program; vii) shoot fresh mass (SFM, g) and viii) root fresh mass (RFM, g): weighed on a semi-analytical scale with a precision of 0.01 g; ix) shoot dry mass (SDM, g) and x) root dry mass (RDM, g): determined by the drying method, using Kraft paper bags, in a forced air circulation oven at a temperature of 65 °C for 72 hours and weighed on a semi-analytical balance with a precision of 0.01 g.

Secondary variables were measured as follows:

i) total dry mass (TDM, g): obtained by adding SDM and RDM; ii) total fresh mass (TFM, g): obtained by summing SFM and RFM; iii) root density (RD, g cm⁻³): expressed as the ratio between RDM and RV; iv) coefficient of robustness (CoR): obtained by the ratio between PH and SD; v) shoot dry mass/root dry mass ratio (SDM/RDM): obtained by the ratio between the two variables; vi) plant height/root length ratio (PH/RL): obtained by the ratio between the two variables; vii) plant height/shoot dry mass ratio (PH/SDM): obtained by the ratio between the two variables; viii) specific length of the root (RL/RDM): obtained by the ratio between the variables; ix) root thickness (RDM/RL): obtained by the ratio between the variables; and x) Dickson's Quality Index (DQI) (Dickson et al., 1960), obtained by the formula (1):

$$DQI = \frac{TDM}{\frac{PH}{SD} + \frac{SDM}{RDM}} \quad (1)$$

Where, DQI: Dickson's quality index; TDM: total dry mass; PH: plant height; SD: stem diameter; SDM: shoot dry mass; RDM: root dry mass.

Validation of the normality of the residues was performed using the Shapiro-Wilk test and an analysis of variance was performed adopting the value of $\alpha = 0.05$ as the significance limit for the F test, using a Generalized Linear Model, according to the statistical model: $Y_{ij} = \mu + T_i + e_{ij}$, where Y_{ij} = observed result for variables; μ = overall mean effect; T_i = treatment effect (CWW doses) and e_{ij} = experimental error. After these analyses, a regression analysis was performed to obtain the optimal dose. The

analyses were performed in the R statistical software (R CORE TEAM 2019) using the 'ExpDes.pt' package (Ferreira et al., 2017).

The variables root length (RL) and root density (RD) had a significant effect with the use of cassava. The other variables did not register significance with the doses of cassava wastewater (CWW) tested (**Tables 2 and 3**).

Observing the root length (RL), there was an effect of the first ($p < 0.05$) and second degree ($p < 0.05$), where the RL was better adjusted to the quadratic model. He noticed that the doses of CWW reduced the RL, with the lowest value recorded at the dose of 15.5 ml L⁻¹. The 0 ml L⁻¹ dose performed better than the other doses (**Figure 2**).

Studying root density (RD), it was found that it had a first ($p < 0.05$) and second degree ($p < 0.05$) effect, better fitting the quadratic model. It was noted that the dose of 13.5 ml L⁻¹ recorded the highest RD with a mean value of 0.08 g cm⁻³ (**Figure 3**).

This reduction (Figure 2) possibly occurred due to the hydrocyanic acid (HCN) present in the CWW. Although plants have greater resistance to cyanide, the compound affects some stages of the Calvin-Benson cycle (sequence of metabolic activities responsible for fixing carbon in photosynthesis) (Hill et al., 2014), harming papaya plant development.

Another possibly contributing factor was the cytogenotoxic effect that CWW caused on initial papaya growth. Ogunyemi et al. (2022) reported that this residue has a cytogenotoxic effect, which generates chromosomal aberrations and distorts plant development. This explains the inhibition of biometric parameters observed in papaya.

Another factor that may have contributed to this reduction in RL is the acidity of the CWW. When discarded in the soil, CWW impairs the balance between nutrients, as it decreases the pH of the soil solution and affects the availability and, consequently, the absorption of nutrients. In addition, CWW increases the salinity of the soil, as it brings a considerable amount of salts into the soil solution (Dantas et al., 2017; Costa et al., 2020).

Another possible cause is the high content of potassium (K) in the composition of CWW. Excess K in

Table 2. Summary of analysis of variance (degrees of freedom and mean squares) of the primary variables plant height (PH), root length (RL), stem diameter (SD), number of leaves (NL), shoot fresh mass (SFM), root fresh mass (RFM), shoot dry mass (SDM), root dry mass (RDM), leaf area (LA), root volume (RV)

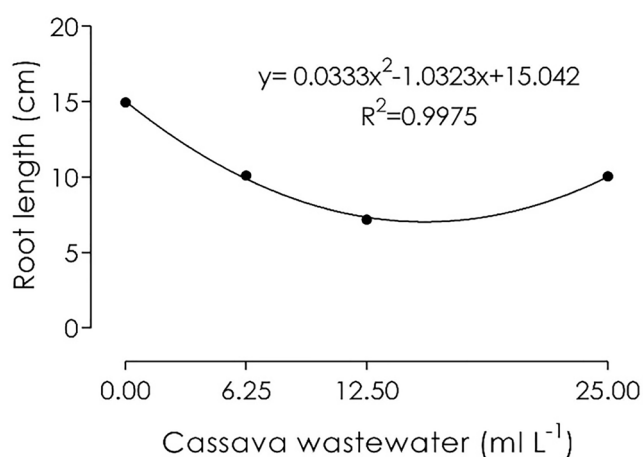
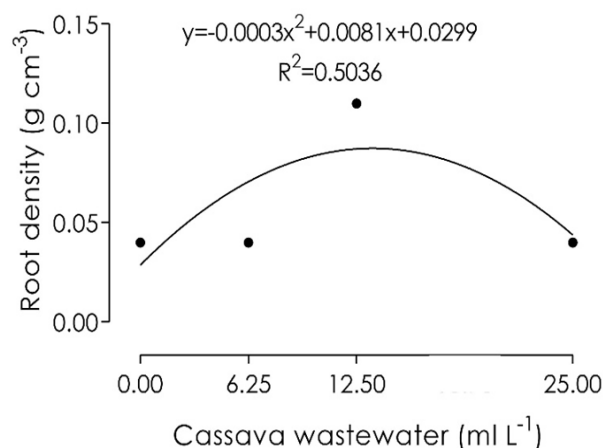
Sources of variation	DF	PH	RL	SD	NL	SFM
Treatment	3	9.21 ^{ns}	41.76 [*]	0.12 ^{ns}	0.47 ^{ns}	0.17 ^{ns}
Residue	12	9.07	8.24	0.14	0.51	0.27
Total	15					
Sources of variation	DF	RFM	LA	SDM	RDM	RV
Treatment	3	0.18 ^{ns}	155.07 ^{ns}	4.7 ^{-3ns}	5.5 ^{-4ns}	0.96 ^{ns}
Residue	12	0.41	459.25	0.01	1.5 ⁻³	1.4
Total	15					

DF: degrees of freedom. *: significant at 5% probability by the F test. ^{ns}: not significant.

Table 3. Summary of analysis of variance (degrees of freedom and mean squares) of secondary variables total dry mass (TDM), total fresh mass (TFM), root density (RD), coefficient of robustness (CoR), shoot dry mass/root system dry mass ratio (SDM/RDM), plant height/root length ratio (PH/RL), plant height/aerial dry mass ratio (PH/SDM), specific length of the root (RL/RDM), root thickness (RDM/RL) and Dickson's Quality Index (DQI)

Sources of variation	DF	TDM	RD	CoR	PH/RL	SDM/RDM
Treatment	3	0.01 ^{ns}	4.5 ^{-3*}	4.05 ^{ns}	0.04 ^{ns}	0.5 ^{ns}
Residue	12	0.02	1.1 ⁻³	4.14	0.06	0.84
Total	15					
Sources of variation	DF	DQI	PH/SDM	RL/RDM	RDM/RL	TFM
Treatment	3	7.8 ^{-4ns}	3530.14 ^{ns}	149569 ^{ns}	1.5 ^{-6ns}	0.63 ^{ns}
Residue	12	2.5 ⁻³	9172.66	297722.8	09 ⁻⁶	1.33
Total	15					

DF: degrees of freedom. *: significant at 5% probability by the F test. ^{ns}: not significant.

**Figure 2.** Root length of papaya formosa seedlings subjected to increasing levels of cassava wastewater.**Figure 3.** Root density of papaya formosa seedlings subjected to increasing levels of cassava wastewater.

the soil solution impairs the absorption of nutrients such as manganese, zinc, iron and calcium, which affected plant development, such as reduced root growth (Taiz et al., 2017; Xie et al., 2021).

This increase in RD (Figure 3) was due to the nutritional contribution of CWW in the substrate solution. However, doses above and 13.5 ml L⁻¹ reduce the development of this variable. The RD is mainly influenced by the root dry mass, which probably increased its values thanks to the nutrient supply provided by the CWW. The factors that reduced the DR with doses above 13.5 ml L⁻¹ may be the same factors that reduced the RL.

Eventually, the increase in soil acidity, imbalance of nutrients in the soil solution, excessive input of K to the

substrate, alteration in the photosynthetic activities of papaya seedlings contributed to the reduction of RD.

It is evident that this effluent has agricultural potential, since it is proven to be rich in nutrients and even caused a certain increase in RD (Figure 4). However, the results obtained in the present research reinforce that the use of this residue must be carried out carefully and after a previous study of its effects, establishing specific doses for the species, as it is a potential pollutant (Carvalho et al., 2021).

In short, it was verified that the CWW doses tested in the present work only influence the RL and RD of the papaya formosa seedlings, and the RL has its development reduced with the use of any dose of CWW

tested in the present research. The RD has an increase in its values, but in large concentrations of CWW, it also has its development reduced.

Conclusions

The use of cassava wastewater negatively influences the root length and positively the root density of formosa papaya seedlings.

A dose of 13.5 ml L⁻¹ of cassava wastewater 28 days after sowing is recommended to increase the root density of formosa papaya seedlings.

As it has a high concentration of nutrients, more research should be carried out on the use of this residue in the production of seedlings of commercial species. As it is an environmental pollutant, its use must be cautious and responsible.

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Conflict of Interest Statement: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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