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Poultry litter biochar rates and incubation times: effects on soil fertility and radish development

Doses de biocarvão de cama de aviário e tempo de incubação: efeitos na fertilidade do solo e desenvolvimento de rabanete

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The effects of biochar on soil nutrients and radish growth were studied.

Soil was incubated with biochar for 30, 60, and 90 days.

Biochar increased soil chemical parameters, which decreased with incubation time.

Biochar stimulated radish growth, improving leaf area, diameters, and fresh biomass.

Abstract _

The objective of this study was to evaluate the effect of increasing rates of poultry litter biochar incubated in soil for different periods on soil fertility attributes and the development of the radish plant. An experiment was carried out in a greenhouse at the Federal University of Campina Grande - UFCG. Treatments consisted of the combination of two factors: four incubation times (0, 30, 60, and 90 days) and four biochar rates (12.5, 25.0, 37.5, 50.0 t ha⁻¹), in three replications, totalizing 48 experimental plots, in a completely randomized design. After the incubation period, soil samples were collected from each experimental unit and chemically characterized. Following this, radish was sown and cultivated for up to 30 days. The agronomic development of radish was evaluated based on the following variables: leaf area; leaf area ratio; specific leaf area; shoot biomass production index; transverse and longitudinal tuber diameters; and fresh and dry tuber biomasses. Under the present experimental conditions, biochar increases soil fertility levels and is able to supply nutrients to the plants in a short period, increasing plant variables.

Key words: Chemical parameters. Organic fertilization. Raphanus sativus L.

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Resumo .

O objetivo deste trabalho foi avaliar o efeito de doses crescentes de biocarvão de cama de aviário, incubado ao solo por diferentes períodos, nos atributos de fertilidade do solo e no desenvolvimento de rabanete. Para tanto, foi realizado um experimento em casa de vegetação, na Universidade Federal de Campina Grande - UFCG, onde os tratamentos resultaram da combinação de dois fatores: 4 tempos de incubação (0, 30, 60 e 90 dias) e 4 doses de biocarvão (12,5, 25,0, 37,5, 50,0 t ha-1) com três repetições, totalizando 48 unidades experimentais, em delineamento inteiramente casualizado. Após o período de incubação, foram coletadas amostras de solo de cada unidade experimental, caracterizadas quimicamente e em seguida o rabanete foi semeado e cultivado por até 30 dias. O desenvolvimento agronômico do rabanete foi avaliado conforme as variáveis: área foliar; razão de área foliar; área foliar específica e índice de produção de biomassa da parte aérea do rabanete; diâmetro transversal e longitudinal do rabanete; biomassa fresca e seca do rabanete. Pelas condições experimentais, concluiu-se que o biocarvão aumentou os níveis de fertilidade do solo e foi capaz de fornecer nutrientes às plantas em um curto período aumentando as variáveis analisadas das plantas.

Palavras-chave: Adubação orgânica. Parâmetros químicos. Raphanus sativus L.

Introduction __

In agricultural activity, the increasing costs of purchase and application of mineral fertilizers have caused producers family farmers, mainly to seek alternative fertilizer options. Thus, several agro-industrial wastes such as cattle manure, swine manure, and poultry litter (Bulgari, Cocetta, Trivellini, Vernieri, & Ferrante, 2015) have been employed to reduce the use of inputs without decreasing the yield or quality of the produced crops, while helping to minimize environmental impacts.

The great expansion of the poultry sector in Brazil has generated excessive amounts of poultry litter, a waste produced in the activity. One of the alternatives for the final destination of this waste is its direct use in agricultural soil as a source of organic matter and nutrients; however, depending on its composition, it can pollute the environment. Another option for using poultry litter as a way to recycle this waste in agriculture is by producing biochar, a material generated from

the process of pyrolysis, described as the thermal decomposition of organic biomass at different temperatures under restricted or zero oxygen (Lehmann & Joseph, 2009).

In general, the use of biochar reduces the pollution potential incurred by the disposal of fresh poultry litter. Additionally, it improves the soil by increasing the levels of important nutrients, increasing the pH, and improving water retention and porosity (Herath, Camps-Arbestain, & Hedley, 2013). As a fertilizer, biochar can increase the yield of short-cycle crops with high nutritional demands, such as radish. However, few studies investigated the benefits of poultry litter biochar regarding soil fertility in short-cycle crops. This is because several factors, including the time of incubation of this material in the soil, influence the process of mineralization of this biochar, which causes the nutrients in its composition to be released into the soil. It is not known exactly how long biochar takes, once applied to the soil, to improve crop production. Jeffery, Coliins and Bailey (2010) applied



biochar to soil and observed that 10 to 20% of the soluble compounds of the material are mineralized to CO_2 and that aromatic and aliphatic compounds can form more complex molecules, increasing resistance to microbial decomposition. According to Sigua et al. (2014), the mineralization of the carbon present in poultry litter biochar applied to two Acrisols resulted in the emission of 12.5 \pm 1.3 and 7.1 \pm 0.7 mg CO_2 /day.

Radish (Raphanus sativus L.) is characterized as one of the shortest-cycle crops among vegetables, taking three to six weeks after sowing to complete its cycle. It is a crop of interest mainly for family farming (Oliveira, Biscaro, Motomiya, Jesus, & Vieira, 2014) and can be intercropped with other species of similar size and cycle (Damasceno, Massaroto, Nascimento, & Munhoz, 2016) and/or cultivated between two other longercycle crops (Cardoso & Hiraki, 2001; Matos, Silva, Lima, Santos, & Dantas, 2015), allowing a quick financial return to farmers. The most widely accepted radish cultivars in Brazil are characterized by their reddish root, white flesh, round shape, and slightly spicy flavor (Hidalgo, Anjos, Freitas, & Cardoso, 2018).

In view of the foregoing, this study was undertaken to evaluate the effect of increasing rates of poultry litter biochar incubated in the soil for different periods on soil fertility attributes and the development of the radish plant.

Materials and Methods __

The experiment was carried out in a greenhouse at the Department of Agricultural Engineering at the Federal University of Campina Grande, located in the state of

Paraiba, Brazil (07°13'11" S; 35°53'31" W). Soil collected at the 0-20 cm depth layer from the *agreste* region of Paraíba was used.

Chemical and physical analysis of the soil samples (Teixeira, Donagemma, Fontana, & Teixeira, 2017) revealed the following attributes: pH (H_2O) = 5.21; EC_{se} = 1.52 dS m⁻¹; Ca = 3.42 cmol_c kg⁻¹; Mg = 3.44 cmol_c kg⁻¹; Na = 1.16 cmol_c kg⁻¹; K = 0.46 cmol_c kg⁻¹; H = 3.43 cmol_c kg⁻¹; Al = 0.20 cmol_c kg⁻¹; organic matter = 6.9 g kg⁻¹; P = 16.2 mg kg⁻¹; clay = 158.5; silt = 120.7; and sand = 720.8 g kg⁻¹.

The biochar used was generated from a pyrolysis process whereby poultry litter was subjected to thermal decomposition at a temperature of 450 $^{\circ}$ C, in the absence of oxygen. After production, biochar samples were oven-dried at 65 \pm 5 $^{\circ}$ C until reaching constant weight, chemically analyzed according to Ministério da Agricultura, Pecuária e Abastecimento [MAPA], (2014), as recommended by ASTM D1762-84 (2007).

The biochar used had the following composition: pH (H_2O) = 9.45; N = 3.45%; P = 7.78%; K = 4.90%; Ca = 6.83%; Mg = 1.34%; S = 0.76%; Fe = 0.46%; Cu = 0.04%; Zn = 0.08%; Mn = 0.09%; B = 0.01%; organic carbon = 39.77%; organic matter = 68.56%; C/N = 11.53%; and CEC = 388.90 mmol_/kg.

This study was laid out in a completely randomized design, in three replicates, with a 4×4 factorial arrangement represented by the following factors: four biochar rates (50, 100, 150, and 200 g per plant, corresponding to 12.5, 25, 37.5, and 50 t ha⁻¹, respectively) and four incubation times (0, 30, 60, and 90 days), totaling 48 experimental units. These biochar rates were determined based on the recommended organic matter (cattle manure) content of 20 m³ ha⁻¹ for most crops. For each



experimental unit, 48 pots were filled with 8 kg of air-dried soil (< 2 mm), which was then mixed with the respective biochar rates. The pots were watered to 80% of field capacity with daily increments based on their weight loss and then closed with plastic film to prevent evaporation, for the incubation periods of 30, 60, and 90 days.

After the incubation period, soil samples were collected from all pots and characterized chemically according to Teixeira et al. (2017). Then, radish (*Raphanus sativus* L. cv. Apolo) was sown directly into the plastic pots, by allocating five seeds equidistantly per experimental unit, at a depth of 3 cm. At 15 days after germination, the pots were thinned, leaving only the most vigorous plant. The experiment was conducted for 30 days.

To assess the effects of incubation times and biochar rates on radish, the following variables were analyzed: leaf area (cm²); leaf area ratio; specific leaf area; shoot biomass production index; transverse and longitudinal tuber diameters (mm); and fresh and dry tuber biomasses (g), according to the equations below:

$$Leaf\ area, cm^2 = L*W*f,$$

where L = leaf length, cm; W = leaf width, cm; and f = correction factor for radish (0.57), dimensionless:

Leaf area ratio,
$$\frac{cm^2}{g} = \frac{TLA}{TDW}$$

where TLA = total leaf area (cm²); and TDW = total dry weight (g);

Specific leaf area,
$$\frac{cm^2}{g} = \frac{LA}{LDW}$$

where LA = leaf area (cm2); DLM = leaf dry weight (g); and

$$SBPI, \frac{g}{g} = \frac{SFW}{TDM},$$

where SBPI = shoot biomass production index, (g/g); SFW = shoot fresh weight (g); and TDM = total dry matter (g).

Prior to analysis of variance (ANOVA), the obtained data were subjected to homogeneity and normality tests using R software (Chambers, 2008). Once the assumptions were met, ANOVA was performed using the F test at the 0.05 probability level. When a significant effect was detected, linear and quadratic polynomial regression analyses were performed using SISVAR statistical software (Ferreira, 2011).

Results and Discussion _

The variation in рH, electrical conductivity, and organic matter, phosphorus, potassium, sodium, calcium, and magnesium contents of the soil followed a quadratic polynomial behavior in response to the applied biochar rates and incubation times (Figure 1). Overall, increasing biochar rates in the soil increased the values of all these parameters. Incubation time caused the pH values of the soil samples to decrease linearly, corroborating Safian, Motaghian and Hosseinpur (2020). The other soil parameters showed higher values in the non-incubated samples compared with those incubated for 30, 60, and 90 days.



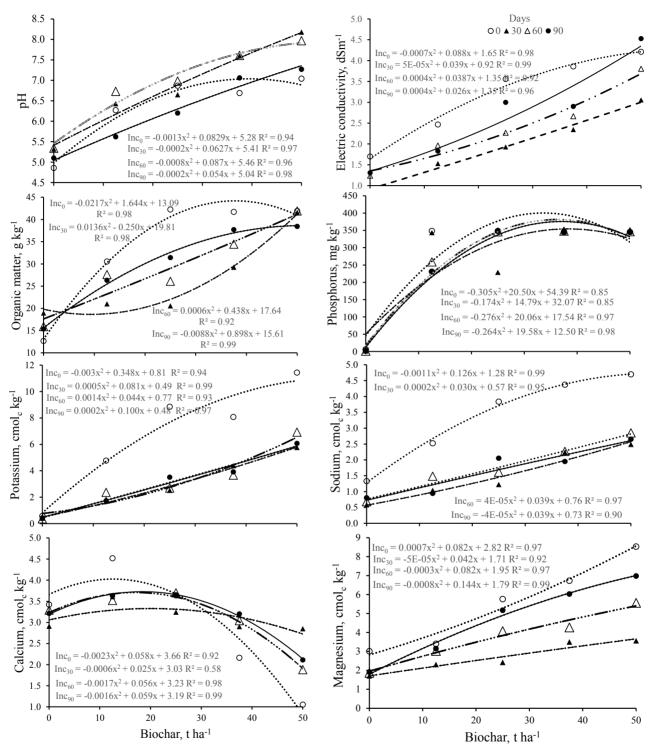


Figure 1. pH, electrical conductivity, organic matter, phosphorus, potassium, sodium, calcium, and magnesium as a function of increasing biochar rates at 0, 30, 60, and 90 days of incubation.



Phosphorus, potassium, and sodium levels practically did not vary with incubation time, as evidenced by their curves almost overlapping each other. This was probably because there was not enough time for biochar to react in the soil, releasing the nutrients from its structure to it. However, available phosphorus in the soil increased with biochar rates up to around 40 t ha-1, corroborating Sousa and Figueiredo (2016) and Safian et al. (2020). The increase in available P content in the soil was due to the high concentration of this nutrient in biochar. Pyrolysis results in loss of carbon (C), hydrogen (H), oxygen (O), and nitrogen (N) by volatilization, thus favoring the P content in biochar. Unlike phosphorus, the concentrations of K and Na increased with the biochar rates up to 50 t ha-1.

X-ray analysis of poultry litter biochar shows the presence of inorganic compounds containing the potassium element, such as sylvite (KCI) and potassium aluminum silicate (KAISiO₄). Feldspar compounds [orthoclase (KAISi₃O₈)] and potassium phosphate [K₂ (HPO₄)] are also identified, confirming the presence of considerable levels of P, K, Ca, and Mg, which influence the concentrations of these elements in the soil. The present X-ray diffraction (XRD) patterns were quite similar to the pattern of biochar derived from chicken litter reported by Chaves et al. (2020).

The Ca content increased gradually with the biochar rates up to around 25 t ha⁻¹, decreasing thereafter. There was also little variation in calcium in the soil over the biochar incubation times. The availability of Ca to the soil is generally linked to the practice of liming on acidic soils, as an effect on limestone.

According to the analysis of XRD patterns of the poultry litter biochar, this is attributed to the presence of calcite in its structure, evidencing the alkaline character of this biochar (Smider & Singh, 2014).

The increasing biochar rates increased the soil Mg content, which also increased with the incubation times of 30, 60, and 90 days.

Overall, it can be stated that the application of poultry litter biochar increased soil fertility levels as a result of the greater availability of exchangeable cations and phosphorus to the soil, thus constituting a viable alternative for short-cycle crops, such as radish. However, several studies have shown that fertility levels can decrease due to the raw material, time, and temperature used in the process of pyrolysis for the production of biochar (Smider & Singh, 2014). In the present study, starting at a rate of 40 t ha⁻¹, poultry litter biochar application to the soil resulted in high pH, especially at the incubation times of 30 and 60 days, possibly impairing the development of radish.

As a result of improved soil fertility, the use of poultry litter biochar significantly influenced leaf area, leaf area ratio, specific leaf area (Table 1), transverse and longitudinal tuber diameters, and fresh tuber biomass (Table 2). Shoot biomass production index was only influenced by incubation time and by the interaction between biochar rates and incubation times. Thus, the results demonstrate the potential of poultry litter biochar to be used as a fertilizer even in very short-cycle crops such as radish.



Table 1 Analysis of variance of leaf area (LA, cm²), leaf area ratio (LAR, cm² g⁻¹), specific leaf area (SLA, cm² g⁻¹), and biomass production index (BPIS, g g⁻¹) of radish shoots as a function of different biochar rates and incubation periods

Source of variation	DF	LA	LAR	SLA	BPIS
Rate (R)	3	13240.78**	2060.38**	40471.05**	0.003 ^{ns}
Linear regression	1	27881.41**	2127.25**	48930.13**	0.001 ^{ns}
Quadratic regression	1	1995.46 ^{ns}	3684.93**	67935.70**	0.006 ^{ns}
Incubation time (I)	3	5466.05*	126.02 ^{ns}	12860.51**	0.01**
Linear regression	1	21243.90**	261.05 ^{ns}	576.10 ^{ns}	0.0002 ^{ns}
Quadratic regression	1	1671.71 ^{ns}	0.72ns	45.36 ^{ns}	0.00002 ^{ns}
R × I interaction	9	10098.87**	434.07*	12052.82**	0.0003*
Error	30	2350.8	170.89	3191.87	0.003
Coefficient of variation, %		14.60	18.23	24.33	16.66

^{*, **} significant at the 0.05 and 0.01 probability levels, respectively; ns - not significant; DF - degrees of freedom.

The effect of biochar rates on the leaf area of the radish plants fitted the linear model (Figure 2A). It is noteworthy that the leaf is the plant part where the biochemical process (photosynthesis) responsible for the production of photoassimilates, which will be sent to the productive organs of the plants, takes place.

Biochar application to the soil without incubation provided the greatest leaf development, with a maximum leaf area of 395.2 cm² achieved at the highest rate of 50 t ha⁻¹. Compared with the smallest leaf area of 211.56 cm2 found at the biochar rate of 12.5 t ha⁻¹, this variable increased by around 86.8%. However, when biochar was incubated in the soil for 60 days, leaf area decreased, with a

reduction of around 21.89% (387.9 to 302.34 cm2) from the lowest to the highest rate of biochar. This last value, which corresponded to 200 g of biochar per plant (highest rate), corroborates Lopes et al. (2019), who studied the application of organic fertilizers and found a leaf area of 304.52 cm² using 200 g of sheep manure per plant. Nonetheless, when the researchers used 200 g of poultry litter per plant, the largest leaf area found was 227 cm². Pereira et al. (2011) analyzed the radish crop fertilized with earthworm humus, at 30 days after transplanting, and found an average leaf area of 450 cm². As illustrated in Figure 2B, incubation time only influenced leaf area at the biochar rate of 12.5 t ha-1, causing it to reach 352.86 cm² at 90 days of incubation.



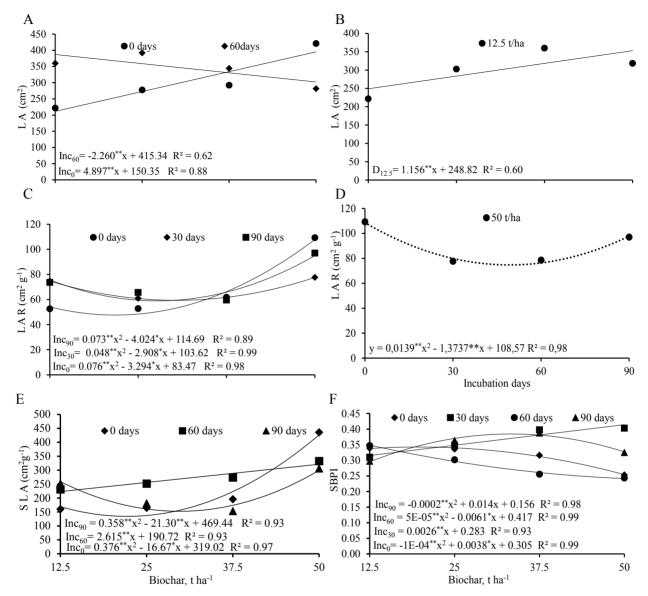


Figure 2. Influence of biochar rates and incubation times on leaf area (LA) (A, B) and leaf area ratio (LAR) (C, D); and influence of biochar rates on the specific leaf area (SLA) (E) and shoot biomass production index (SBPI) of radish (F).

The morphophysiological component that expresses the leaf area useful for photosynthesis and the total dry weight of radish, i.e., leaf area ratio (Figure 2C), was significantly affected by the interaction between incubation time (only at 0, 30, and 90 days) and biochar rates. These data fitted the

quadratic model, with the maximum leaf area ratios found at the rate of 50 t ha⁻¹: 108.77, 78.22, and 95.99 cm² g⁻¹ at 0, 30, and 90 days of incubation, respectively. The interaction between biochar rates and incubation times (Figure 2D) only affected this variable at the rate of 50 t ha⁻¹, with a maximum point of



108.57 cm² g⁻¹ seen in the plants that received biochar without incubation.

Non-incubated biochar provided the largest leaf area ratio, thus influencing photosynthesis. Specific leaf area, which expresses the photosynthetically active area of radish relative to its leaf dry weight, was influenced by the incubation time vs. biochar rate interaction (Figure 2E). This variable showed linear and quadratic fits of the data, with 426.57, 321.45, and 299.39 cm² g⁻¹ found at the incubation times of 0, 60, and 90 days, respectively, at the rate of 50 t ha⁻¹. It is worth mentioning that the largest specific leaf area was attained without incubating biochar, suggesting that it does not require incubation to release the nutrients needed by radish. Lower values were observed by Matos et al. (2015), who studied the effects of wastewater and a drip application system and found a specific leaf area of 311.8 cm² g⁻¹.

There was a significant biochar rate vs. incubation time interaction effect (Figure 2F) on shoot biomass production index, whose data showed quadratic and linear fits with maximum points of 0.34, 0.41, 0.35, and 0.40 at the biochar rates of 19, 50, 12.5, and 35 t ha⁻¹, respectively. By analyzing the data, we observe that the biochar rate of 50 t ha⁻¹ provided the largest accumulation of shoot dry biomass at the incubation time of 30 days, probably due to the concentration of nutrients that biochar provided during incubation.

The transverse and longitudinal diameters of the tuber were influenced by the interaction between biochar rates and incubation times, with a significant effect at 1% probability. However, biochar rates in isolation affected only the fresh biomass of the tuber (Table 2).

Table 2

Analysis of variance of transverse and longitudinal tuber diameters and fresh and dry biomasses of radish as a function of biochar rates and incubation periods

Source of variation	DF -	Radish tube diameter		Biomass	
		Transverse	Longitudinal	Fresh	Dry
Rate (R)	3	46.26**	188.32**	333.21*	0.70 ^{ns}
Linear regression	1	0.68 ^{ns}	389.21**	146.39 ^{ns}	0.03 ^{ns}
Quadratic regression	1	134.23**	174.16*	767.84**	2.03*
Incubation time (I)	3	41.00**	121.79*	172.85 ^{ns}	0.26 ^{ns}
Linear regression	1	4.78 ^{ns}	119.53 ^{ns}	355.17**	0.20 ^{ns}
Quadratic regression	1	109.05**	239.10**	162.14 ^{ns}	0.25 ^{ns}
R × I interaction	9	50.77**	386.52**	74.00 ^{ns}	0.16 ^{ns}
Error	30	4.77	22.42	87.74	0.34
Coefficient of variation, %		5.40	8.20	21.70	21.50

^{*, **} significant at the 0.05 and 0.01 probability levels, respectively; ns - not significant; DF - degrees of freedom.



The transverse diameter of the tuber (Figure 3A) fitted a quadratic model, with maximum values of 45.29, 43.87, 46.06, and 42.43 mm at the maximum biochar rates of 31.55, 32.46, 50, and 30.22 t ha⁻¹, respectively, at the best incubation time of 60 days. In the interaction between biochar rates and incubation time on transverse diameter (Figure 3B), 37.5 t ha⁻¹ of biochar without incubation resulted in the largest diameter of 45.9 mm. This value is similar to the 41.57 and 44.34 mm/ plant found by Cecconello and Centeno (2016), who used vermicompost of plant residues.

Vitti, Vidal, Morselli and Faria (2007) reported a transverse diameter of 37 mm/plant with the application of 20 g of cattle manure in an experiment with organic fertilizers on radish. Likewise, Rodrigues, Reis and Reis (2013) found a root diameter of around 31.7 mm/plant using chicken manure; Maia et al. (2018) investigated the yield of radish fertilized with different rates of cattle manure and described an average tuber diameter of 30.6 mm; and Rodriguez et al. (2017) found average diameters between 10.8 and 17.7 mm/plant in tubers fertilized with cattle manure vermicompost. These values were lower than those found in the present study, confirming the efficiency of biochar as a fertilizer.

Longitudinal diameter (Figure 3C) was significantly affected by the incubation time

vs. biochar rate interaction. The data fitted a quadratic model, with a maximum diameter of 92.12 mm achieved at the rate of 12.5 t ha⁻¹ and incubation time of 30 days. The analysis of the interaction between biochar rates and incubation times (Figure 3D) revealed that the greatest diameter of 84.9 mm was achieved with an incubation time of 42 days, using the biochar rate of 12.5 t ha⁻¹. This diameter was larger than the 23.62 to 24.34 m observed by Vitti et al. (2007) with the application of 5 to 50 g of cattle manure vermicompost/6 kg of soil. According to these authors, root diameter stagnated with the application of vermicompost levels above 20 g.

The higher values of some variables in plants cultivated with non-incubated biochar can be explained by the greater availability of chemical elements such as phosphorus, potassium, magnesium, calcium, and organic carbon in the soil provided by biochar, as can be seen in Figure 1.

Biochar rates significantly affected the fresh biomass of the radish tuber, which responded quadratically, with a maximum point of 48.4 g at 33.69 t ha⁻¹ of biochar (Figure 4). This biomass weight was higher than the 10.46 g to 17.78 g observed by Vitti et al. (2007) following the application of 5 to 50 g of cattle manure vermicompost/6 kg of soil.



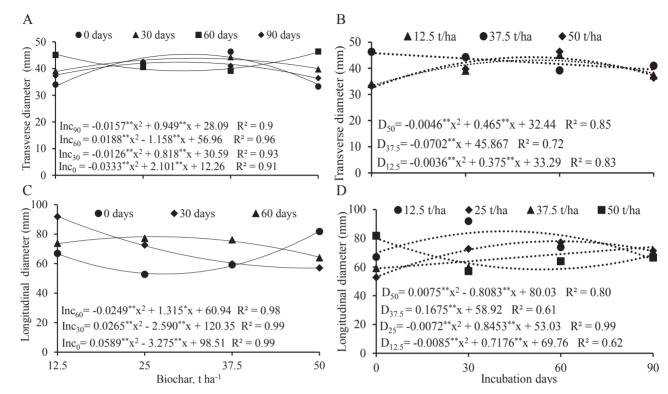


Figure 3. Influence of biochar rates (A, C) and incubation times (B, D) on the transverse and longitudinal tuber diameters of the radish crop.

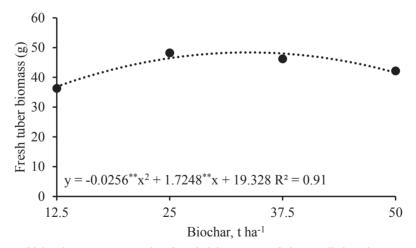


Figure 4. Influence of biochar rates on the fresh biomass of the radish tuber.



Rodriguez et al. (2017) found radish fresh root biomass values of 10.5, 11.6, and 15.65 g with the application of increasing rates of cattle manure vermicompost. Dourado et al. (2012) observed an influence of caprine manure on radish production, reporting that the rate of 7.65 kg m⁻² provided the best result for shoot fresh weight, of 39.26 g. As stated by Reis, Rodrigues and Reis (2012), the use of chicken manure also yielded positive responses in radish root biomass production. However, superior results of 15.15 to 54.04 g were described by Cecconello and Centeno (2016), who used vermicompost from plant residues.

According to Bonela, Santos, Sobrinho, & Costa Gomes (2017), the growth and development of radish roots are probably related to the biochar content applied to the soil and mainly to its physical characteristics, e.g., higher levels of sand, which directly influence porosity, providing better yields.

In general, the decrease in some variables of the radish plant with increasing rates of poultry litter biochar is likely related to the saline effect caused by the presence of salts that accumulated in the soil, provided by the composition of this biochar. This greater salinity in the soil increased electrical conductivity, but this variable decreased with the biochar incubation time.

Radish is a high-moisture food, with an average water content of 95.6 g per 100 g of tuber. Despite the different weights shown by the tuber in its fresh state, after drying, the loss of water resulted in very similar weights. This probably explains the non-significant effect of biochar rates on the dry tuber biomass in spite of the significant effect on fresh biomass.

Conclusion

The pH, electrical conductivity, nutrient contents (mainly phosphorus, potassium, and magnesium), and organic matter in the soil samples treated with biochar increased with increasing rates of the product. In addition, in all of these samples, these parameters decreased with incubation time.

Poultry litter biochar was able to supply nutrients to the plants in a short period, providing overall increases in leaf area, leaf area ratio, specific leaf area, and transverse and longitudinal tuber diameters. The rate of 33.69 t ha⁻¹ resulted in the largest radish fresh biomass production, of 48.4 g.

The poultry litter biochar rates of 25 to 37.5 t ha⁻¹ were the range that provided the best results of the analyzed plant variables, within an incubation period of 0 to 30 days.

Poultry litter biochar can be considered a viable alternative source of organic fertilizer.

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