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Growth of pecan rootstocks with the use of liquid humus in an organic production system

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Abstract

Pecan farming is a long-term investment which needs quality seedlings to establish an orchard. Pecan rootstocks must have a vigorous radicular system to form quality seedlings. Liquid humus, which acts as an agent that promotes their growth and development, is a bioinput that is specially used in organic production systems. This study aimed at evaluating effects of different frequencies of liquid humus application on pecan rootstock growth and development in an organic system. Liquid humus at 30% was applied every 7, 14, 21 and 28 days. The control treatment consisted of water application. The experiment was carried out with 'Barton' rootstocks in a greenhouse at the Embrapa Clima Temperado located in Pelotas, RS, Brazil. The following variables were evaluated 280 and 480 days after transplantation: plant height; stem diameter; leaf area; fresh and dry masses of the aerial part; main and secondary root lengths; fresh and dry masses of roots; fresh and dry masses of the main root; fresh and dry masses of secondary roots; and the Dickson Quality Index. Liquid humus increased secondary root length of pecan rootstocks, mainly when it was applied every seven days. Thus, the bioinput may be used for this purpose.

Keywords: Biofertilizer, Carya illinoinensis, radicular development, sexual propagation

Introduction

Since pecan [*Carya illinoinensis* (Wangenh.) K. Koch] farming has gained ground in Brazil, mainly in Rio Grande do Sul (RS) state (Bilharva et al., 2018), there has been increasing search for seedlings. In this crop, seedling development is laborious and requires high investment when orchards are implanted (Bilharva et al., 2018; Hellwig, 2020). The traditional method of grafted pecan seedlings comprises two phases, i. e., sexual propagation (by seeds), which is used for producing rootstocks, and asexual propagation, which consists in grafting the crown cultivar (Poletto et al., 2015; Casales et al., 2018; Zhu et al., 2020). However, rootstocks derived from pecan seeds not only exhibit differences in plant growth and performance but also require a lengthy period to grow and end the seedling production process (Warren, 2015; Vahdatl et al., 2020; Hilgert et al., 2020).

High cost of seedlings is due to slow initial growth of rootstocks, before they can be grafted, and to their maintenance in nurseries, before they are sold. Therefore, appropriate management of seedling production – which starts with rootstock development and proceeds to grafting and graft development – is fundamental and must be addressed by studies of different techniques.

Thus, to favor root formation and, consequently, development of aerial parts of pecan trees, phytoregulators may be used as synthetic agents (Casales et al., 2018). However, in organic production systems, little farms and/or to grow few seedlings, this type of technology is not allowed and may also be costly. As a result, liquid humus, which is the infusion of solid humus in water as the solvent (Zandonadi et al., 2007; Schiedeck et al., 2008; Souza et al., 2018), has been investigated to promote plant growth and development.

Application of liquid humus to plants may lead to increase in their tolerance to pathogens, biological

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nitrogen fixation and nutrition, amount of carbon in the soil and number of microorganisms; thus, the environment becomes more favorable to plant growth and development (Román et al., 2013). However, information on the use of liquid humus as a biostimulant is scarce, since studies have just started, mainly on pecan trees. Therefore, this study aimed at evaluating effects of different frequencies of liquid humus application on pecan rootstock growth and development in an organic system.

Material and methods

The experiment was carried out in 2018, 2019 and 2020 with rootstocks sown in 2018 at the Embrapa Clima Temperado, Estação Experimental Cascata (EEC), in Pelotas, RS, Brazil (latitude 31°37'9" S; longitude 52°31'33" W; altitude of 170 m). In May 2018, seeds of the cultivar Barton were scarified by an angle grinder, stratified in wet sand boxes and taken to a cold chamber at 3°C for 95 days. On August 22nd, 2018, after they broke dormancy, seeds were placed on sand beds in a greenhouse so that they could germinate and emerge. On October 15th, 2018, fifty-three days after sowing, seedlings were transplanted to polyethylene containers (49.0 x 10.0 cm) with the comercial substrate Ecocitrus®.

The experiment had a completely randomized design with 5 replicates. Every experimental unit was composed of five seedlings. Treatments consisted in liquid humus application at different frequencies, i. e., every 7, 14, 21 and 28 days, besides the control (no bioinput; just water). The dose of liquid humus was 100mL at 30%, in agreement with results published by (Zandonadi et al., 2016) and (Souza et al., 2018). Besides liquid humus application, rootstocks were irrigated with water, depending on their water needs. In the 2018/2019developmental season, 26, 13, 9 and 7 applications were carried out every 7, 14, 21 and 28 days, respectively, while in the 2019/2020 season, 20, 10, 7 and 5 applications were conducted every 7, 14, 21 and 28 days, respectively.

Solid humus derives from vermicompost from cattle manure cultured by *Eisenia fetida* earthworms. It was prepared in a plastic container, where 6 kg solid humus (at 50% moisture) was placed in a voile bag and made up to 10 liters of liquid humus with water, depending on the application frequency. The infusion was prepared 24 hours before its application by an aeration pump which enables air circulation and release of particles of interest.

Growth was evaluated 280 days (25 rootstocks) and 480 days (25 rootstocks) after transplants in July 2019 and February 2020, respectively. Height of the aerial part and length of the primary and secondary radicular system were measured by ametal tape measure (cm). Stem diameter was measured by a digital pachymeter (mm) three centimeters above the substrate. Leaf area was read by a LI-COR® - LI-300C area meter (cm^2). Fresh and dry matter of the aerial part and of the radicular system (primary and secondary roots, separately) was determined by a Bioscale electronic scale (0.1g). Material was dried in a forced air circulation oven at 65o C, up to constant weight.

Taking into consideration indicators of dry mass of the aerial part and roots, besides total dry mass, height and base diameter, rootstock quality was evaluated by the Dickson Quality Index (DQI), proposed by (Dickson et al., 1960). DQI = TDM/(H/BD) + (DMAP/DMR), where TDM is total dry mass (g); H is plant height (cm); BD is base diameter (mm); DMAP is dry matter of the aerial part (g); and DMR is dry matter of roots (g). Resulting data were subject to the Analysis of Variance and means were compared by the Tukey's Test at 5% probability by the SISVAR statistical program version 5.6 (Ferreira, 2014).

Results and discussion

Length of the main root was not affected by the frequency of liquid humus application after 280 and 480 days (**Table 1**). Pecan radicular characteristics show a taproot and deep rooting, which not only ensure search for soil humidity that is needed in dry periods but also enable pecan trees to survive in semiarid regions (Sparks, 2005).

Different results of lengths of secondary roots were found 280 and 480 days after transplant (Table 1): 280 days after transplant, applications every 7, 14 and 28 days led to longer secondary roots than the ones of the control while 480 days after transplant, applications every 7 and 21 days led to longer ones, by comparison with the control. It should be highlighted that applications every 7 days contributed to secondary root growth in both periods under evaluation, a fact that presupposes that the most intensive application is more efficient than the others to form secondary roots. The use of liquid humus influences seedling development due to its humic and fulvic acids (Rose et al., 2014), which may bring physiological benefits to plants as growth promoters (Aguiar et al., 2013) that help better rooting to take place (Nardi et al, 2002). The radicular system is an important parameter to produce seedlings since long roots help to increase the capacity that plants have to absorb water and nutrients and may lead to better development and survival after seedlings are planted in the field (Souza et al., 2018).

In general, liquid humus application was efficient

Table 1. Main root length, secondary root length, fresh mass of roots, dry mass of roots of pecan rootstocks subject to different frequencies of liquid humus 280 and 480 days after seedling transplant

Treatments	Main root length (cm)		Secondary root length (cm)		fresh mass of roots (g)		Dry mass of roots (g)	
	280 days after seedling transplant							
Control	51.52	ns	27.44	c	48.56	ab	23.68	ns
7 days	56.82		31.72	ab	46.54	b	22.15	
14 days	57.52		32.72	ab	49.72	ab	26.92	
21 days	47.36		28.42	bc	48.24	ab	23.66	
28 days	54.33		34.11	a	58.06	a	25.88	
Linear	ns		ns		ns		ns	
Quadratic	ns		ns		ns		ns	
CV (%)	10.89		8.12		17.15		18.83	
	480 days after seedling transplant							
Control	51.51	ns	27.47	b	41.88	ns	21.64	ns
7 days	56.61		33.53	a	48.40		18.75	
14 days	56.60		32.55	ab	44.90		20.88	
21 days	52.11		33.11	a	53.06		24.04	
28 days	52.08		30.39	ab	45.90		19.13	
Linear	ns		ns		ns		ns	
Quadratic	ns		ns		ns		ns	
CV (%)	10.17		7.27		18.6		18.97	

Means followed by different letters differ among themselves by the Tukey's test at 5% probability. ns = not significant.

at promoting secondary root growth by comparison with the control treatment. Results corroborate the ones found by (Zandonadi, 2015) who reported that liquid humus has molecules that are similar to auxin, a plant hormone that contribute to vigorous rooting, with a high number of absorbent hairs and secondary roots in strawberry. Root development on the surface layer of the soil ensures nutrient absorption from the richest layer and, when the lower profile of the soil is saturated, it enables root aeration and water and nutrient absorption (Sparks, 2005). Good development of secondary roots may be beneficial to seedlings and their water and nutrient absorption, mainly when there is nutrient deficiency and water shortage. Lateral roots develop horizontally and usually keep superficial (Casales et al., 2018), a fact that ends up valorizing the emission of secondary roots to capture water and nutrients in the soil.

Although liquid humus has led to development of secondary roots, the period under investigation was not enough to determine significant changes in fresh and dry masses of roots. Regarding fresh mass of roots, 280 days after transplant, liquid humus application every 28 days exhibited better results than its application every 7 days, but it did not differ from the other treatments (Table 1). No significant difference in values of dry mass of roots was found among treatments. Even though secondary roots stood out throughout liquid humus application, it was not enough to show any difference in mass because they are fine radicles composed of water and mineral ions. Thus, fresh and dry masses may not be safe criteria to show effects of liquid humus on pecan seedlings. (Ribeiro et al., 2016) did not find any increase in dry and fresh matter, nor in mean number and length of roots when there was increase in humus concentration. However, (Souza et al., 2018) observed that liquid humus at 30% led to good development of fig seedlings. Therefore, since it seems clear that species give specific responses to liquid humus, further studies should address other concentrations and frequencies to better understand its effect on pecan trees.

In general, 480 days after transplant, the frequency of liquid humus application did not interfere with the volume of roots expressed as fresh mass of the main root and dry mass of secondary roots (**Table 2**). Fresh mass and length of the main root were not affected by the frequency of liquid humus application. However, when it was applied every 21 days, dry mass of the main root was higher, by comparison with its application every 28 days (Table 2). Concerning fresh mass of secondary roots, liquid humus application every 7 days led to higher means than the ones found when it was applied every 21 and 28 days (Table 2), but there was no significant difference by comparison with application every 14 days and the control. In contrast, dry mass of secondary roots did not show any significant difference among treatments. The result shows that the highest frequency of application, i. e., every 7 days, may lead to better development of secondary roots, since it was efficient at producing the highest fresh mass of secondary roots, which agrees with the variable length of secondary roots. (Santos et al., 2014) found positive radicular effect on development of plants subject to application of humic

Table 2. Fresh and dry masses of main and secondary roots of pecan trees subject to different frequencies of liquid humus 480 days after seedling transplant

Treatments	Fresh main root (g)		Main root dry mass (g)		Fresh mass of secondary roots (g)		Dry mass of secondary roots (g)	
Control	46.49	ns	18.56	ab	5.18	ab	2.08	ns
7 days	44.71		17.80	ab	5.68	a	1.95	
14 days	40.00		18.74	ab	4.90	ab	2.14	
21 days	48.49		21.31	a	3.54	bc	2.19	
28 days	42.12		16.56	b	2.60	c	1.33	
Linear	ns		ns		ns		ns	
Quadratic	ns		ns		ns		ns	
CV (%)	18.99		18.74		33.94		36.45	

Means followed by different letters differ among themselves by the Tukey's test at 5% probability. ns = not significant.

substances.

Frequency of liquid humus application 280 and 480 days after seedling transplant did not influence plant height, stem diameter, leaf area and fresh mass of the aerial part significantly (**Table 3**), except dry mass of the aerial part 480 days after transplant; in this case, application every 21 days stood out by comparison with the control. It should be mentioned that more radicular development could have taken place as evaluation time advanced. It would result in compensation of growth of the aerial part. Modifications in the radicular system of pecan trees due to chemical fertilization do not affect growth of the aerial part but increase root ramification and total root growth (Casales et al., 2018).

Regarding the analysis of DQI carried out 280 days after transplant, the treatment in which liquid humus was applied every 14 days was statistically better, but it did not differ from treatments whose applications were conducted every 21 and 28 days (Table 3). It shows that the use of the compost at these frequencies improves standards of rootstocks, as a whole, in their first year. In evaluations carried out in the second season, 480 days after transplant, there was no significant difference among treatments. It implies that, in this season, plants balanced

Table 3. Plant height, stem diameter, leaf area, fresh mass of the aerial part, dry mass of the aerial part and means of Dickson Quality Indexes (DQI) in pecan rootstocks subject to different frequencies of liquid humus 280 and 480 days after seedling transplant

Treatments	Plant height (cm)		Stem diameter (mm)		Área foliar (cm²)		Fresh mass of the aerial part (g)		Dry mass of the aerial part (g)		IQD	
280 days after seedling transplant												
Control	24.98	ns	5.25	ns	524.88	ns	11.04	ns	5.26	ns	5.58	b
7 days	25.22		5.24		546.24		8.84		4.88		5.91	b
14 days	26.14		5.56		611.16		9.24		5.93		8.12	a
21 days	26.19		5.40		581.92		9.88		5.93		6.25	ab
28 days	25.58		5.50		497.84		9.04		5.28		6.82	ab
Linear	ns		ns		ns		ns		ns		ns	
Quadratic	ns		ns		ns		ns		ns		ns	
CV (%)	7.43		6.45		14.57		23.02		20.81		15.73	
480 days after seedling transplant												
Control	31.31	ns	6.04	ns	437.68	ns	16.52	ns	6.78	b	5.16	ns
7 days	34.81		6.39		414.34		17.27		8.82	ab	5.05	
14 days	31.01		5.78		424.10		15.38		7.68	ab	5.43	
21 days	33.49		6.38		540.84		21.22		10.04	a	5.87	
28 days	34.00		5.88		388.89		15.74		7.55	ab	3.87	
Linear	ns		ns		ns		ns		ns		ns	
Quadratic	ns		ns		ns		ns		ns		ns	
CV (%)	9.19		6.15		18.41		19.54		18.58		21.71	

Means followed by different letters differ among themselves by the Tukey's test at 5% probability. ns = not significant.

their growth, regardless of liquid humus application.

In general, frequency of liquid humus application did not improve most variables under evaluation by comparison with the control but it should be highlighted that one of the benefits of liquid humus related to lateral root growth was reached by results of length of secondary roots. Since secondary roots are responsible for most nutrient and water absorption, producing seedlings without them may lead to decrease and delay in plant growth after cultivation. (Du Jardin, 2015) reported that positive effects of humic substances (soluble fractions of humic and fulvic acids) as growth promoters are subject to variation, sometimes inconsistent, because results depend on the interaction among organic matter, microorganisms and plant roots. A recent meta-analysis of random effect of humic substances applied to plants showed that the variability of effects is due to humus origin, environmental conditions, plants, doses and modes of humic substance application (Rose et al., 2014).

Although further studies are needed to know the relation between liquid humus and aerial parts of plants, the data analysis of this study showed that beneficial effects on seedling development tend to occur when liquid humus is applied every 7 days, mainly secondary root growth, which may lead to plants with better quality.

Conclusions

Liquid humus application every 7 days is efficient at growth of 'Barton' pecan secondary roots up to 480 days after seedling transplant.

Frequency of liquid humus application did not interfere with the development of the aerial part, height and diameter of rootstocks in the period under evaluation.

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