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INDOLEBUTYRIC ACID (IBA) AND SUBSTRATE IN ROOTING AND DEVELOPMENT OF BRAZILIAN PEPPERTREE CUTTINGS

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Keywords: Anacardiaceae; Native specie; Phyto regulators; Propagation materials; *Schinus terebinthifolia.*

Abstract

This work aimed to evaluate the development of seedlings by cuttings Brazilian peppertree cuttings submitted to different treatments and substrates. For that we used a randomized experimental design in a 2x4 factorial scheme, consisting of the use (3.000 mg.L^{-1}) or not of IBA associated with different types of substrate: vermiculite (V); carbonized rice husk with vermicompost (CV); carbonized rice husk with vermicompost and sand (CVS); Turfa Fértil® commercial substrate (CS), in four replicates per treatment, each consisting of 10 cuttings. After 90 days, the following variables were evaluated: percentage of rooted cuttings, length of the largest root, number of leaves over 1 cm in length, and dry root weight. The Generalized Linear Model (GLM) was used with $p < 0.05$ of significance in the data analysis. The use of IBA in rooting Brazilian peppertree cuttings is recommended. CVS is the substrate with the best interactions, and substrates like CS should be avoided.

ÁCIDO INDOLBUTÍRICO (AIB) E SUBSTRATOS NO DESENVOLVIMENTO DE ESTACAS DE PIMENTA ROSA.

Palavras-chave: Anacardiaceae; Espécie nativa; Fito reguladores; Propagação; *Schinus terebinthifolia.*

Resumo

O trabalho teve como objetivo avaliar o desenvolvimento de mudas por estacas de aroeira-vermelha submetidas a diferentes tratamentos e substratos. Foi utilizado delineamento experimental inteiramente casualizado em um esquema fatorial 2x4, constituído no uso (3.000 mg.L-1) ou não de AIB associado à diferentes tipos de substrato: vermiculita (V); casca de arroz carbonizada com vermicomposto (C); casca de arroz carbonizada com vermicomposto e areia (CA); substrato comercial Turfa Fértil® (SC), em quatro repetições por tratamento, cada uma composta por 10 estacas. Após 90 dias foram avaliadas as seguintes variáveis: percentual de estacas enraizadas, comprimento da maior raiz, número de folhas ($\gtrsim$ 1 cm de comprimento) e massa seca de raízes. Na análise dos dados foi utilizado o Modelo Generalizado Linear (GLM) ($p < 0{,}05$ de significância). O uso de AIB foi benéfico para o enraizamento de estacas de pimenta rosa. Sendo CA o substrato com melhores interações, e devendo-se evitar substratos semelhantes ao SC.

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INTRODUCTION

Schinus terebinthifolia Raddi (Anacardiaceae), the Brazilian peppertree, which can be found in Brazil, Argentina, Paraguay, Uruguay, and Venezuela (SOBRAL, 2006), produces fruits that have been known as rose pepper. Since they have been increasingly used as condiments in national and international markets. In Brazil, the species has been explored by extractivism, rather than being cultivated (LENZI; ORTH, 2004). It is characterized by reproductive precocity, i. e., it may bloom and fructify in the first year; thus, it is highly recommended for agroforestry systems, recovery of degraded areas, and urban reforestation (MAZZA et al., 2011). High-quality seedlings must be available to ensure satisfactory cultivation and management, while studies of its reproduction are needed to improve its efficiency in terms of production.

Cutting is a method of vegetative propagation which uses segments of a matrix plant (stems, leaves, or roots) to form complete plants, which depend on adventitious root formation on the material being used. This simple method is worth using either seed shortage and infeasibility or genotype selection and multiplication to maintain desirable traits in production systems (XAVIER, 2013). Among propagation methods that can be used with Brazilian peppertree, cutting is especially recommended due to the functionally dioecious reproduction of the species (CESÁRIO; GAGLIONE, 2008), since it keeps an adequate relation between female and male plants to be cultivated in planned production systems (i.e., MARTINS et al., 2019). In addition, certain characteristics, such as productivity and fruit quality, ensure the implementation of homogeneous commercial orchards (SOUZA et al., 2018).

Success in the method development depends on regenerative capacity that tissues must emit adventitious roots and bud shooting (SILVA et al., 2011). To stimulate rooting, substances with properties that regulate plant growth, such as auxins, are used, since they are responsible for root formation in plants and naturally synthesized in apical buds and new leaves (XAVIER, 2013). Their cutting application aims to accelerate root initiation, increase the percentage of rooted cuttings, the number, and quality of resulting roots, and rooting uniformity (FACHINELLO et al., 2005).

Indolebutyric acid (IBA) is a synthetic auxin that has been widely used as a rooting stimulator (TAIZ; ZEIGER, 2004), whose activity is localized and persistent. Tests with different dosages of concentration have shown that the product does not have a toxic effect (MIRANDA et al., 2004). Dosages between 2,000 and 4,000 mg.L^{-1} are usually recommended and evaluated for rooting and blooming of woody cuttings of arboreal species (PEREIRA et al., 2017, SOUZA et al. 2018). In the case of the Brazilian peppertree, Holanda et al. (2012) evaluated the effect of different IBA concentrations (0, 2,500, and 5,000 mg.L^{-1}) and showed that the concentration of 2,500 mg L^{-1} exhibited the best results for rooting.

The substrate is also especially important to the rooting process of plant cuttings because it provides support, enables root aeration, and offers conditions of humidity and nutrition that are needed for radicular system growth. Since there are some types of substrates and different responses are given to them – depending on the plant –, distinct substrates should be assessed for the species of interest (XAVIER, 2013). Substrates that have been successfully used for rooting woody cuttings include sand (BRANDÃO et al., 2020; CUNHA et al., 2015), commercial ones (CUNHA et al., 2015), vermiculite (DUARTE et al., 2020), and vermicompost (ANTUNES et al., 2016). They may be used either pure or mixed in different compositions.

The AIB effect as a rooting stimulator depends on the species and substrate understudy, as all factors interact (SCHUCH et al., 2007). In addition, substrates with high moisture-holding capacity and low-temperature variation show impressive survival results for IBA-treated cuttings (SCHUCH et al., 2007). Although the Brazilian pepper tree doesn't present any difficulty in establishing itself in a natural environment, its development in greenhouses lacks studies to optimize the propagation technique to maintain desirable characteristics both for crops that aim to produce rose pepper and to produce seedlings for restoration. In this sense, the work aimed to evaluate the development of cuttings of the species submitted to treatments with and without IBA in different substrates.

MATERIAL AND METHODS

The study was conducted at the head office of Embrapa Clima Temperado, in Pelotas, Rio Grande do Sul state, in the south of Brazil (31°40`49``S and 52°26`23``W). The climate in the region has been defined as the Cfa type, i. e., humid temperate with hot summers and no dry season (ALVARES et al., 2013). Woody cuttings of 20 Brazilian peppertree adult matrices were randomly collected at end of August 2018 on the edges of fragments of secondary forests. They were 20 cm in length and their diameters in the basal extremity ranged between 0.6 and 0.8 cm. They were extracted from the median portion of young twigs at least 10 cm from the apical extremity, as Ferreira et al. (2003) recommended.

The experiment had a completely randomized design (CRD) in a 2x4 factorial scheme, in the presence (3.000 mg.L-1)) and absence of IBA associated with different types of substrate: i) V - vermiculite; ii) CV - carbonized rice husk with vermicompost (1:1); iii) CVS - carbonized rice husk with vermicompost and sand (1:1:1); vi) CS - Turfa Fértil® commercial substrate (SPP G.P.H. PLUS - composed of peat and carbonized rice husk and enriched with N (0.04%), P_2O_5 (0.04%), K2O (0.05%) and calcite limestone (1.5%), in agreement with the manufacturer's instructions. Four replicates were installed in every treatment; they consisted of 10 cuttings (n = 40 cutting/treatment). In the case of treatments with immersion in IBA, cuttings had 4 cm of their basal extremities immersed in the solution for 10 seconds. Later, they were planted in the substrates under investigation. Cuttings were planted on expanded polystyrene trays in the float system and kept in a greenhouse for 90 days.

The following variables were analyzed 90 days later: percentage of rooted cuttings, length of the longest root, number of leaves longer than 1 cm, root dry mass, presence, and number of buds (leaf emergences and leaves that are less than 1 cm long). Leaf length, leaf emergences, and the longest root were determined by a graduated ruler (cm) while root dry mass was found in agreement with Amaral et al. (2017).

Data were analyzed on R 3.6.1 platform (R CORE TEAM, 2019) by descriptive statistics and the Generalized Linear Model (GLM) at $p < 0.05$ significance to verify the interaction among factors and their contrasts for every variable. The test of overdispersion had been conducted before to analyze which distribution was the most adequate one for every variable. As a result, it was determined that binomial, Gaussian, and Poisson distributions should be used for binary, quantitative continuous, and quantitative discrete variables, respectively.

RESULTS AND DISCUSSION

The results showed that the use of Indolebutyric acid (IBA) was positive and statistically significant (GLM; $P < 0.05$), except with bud presence (Table 1). The results showed that the use of Indolebutyric acid (IBA) was positive and statistically significant (GLM; $P < 0.05$), except with bud presence (Table 1). These results infer that the use of IBA is advantageous because it accelerates the process of seedling development and enables greater efficiency in production both for pink pepper producers who wish to select matrices through the stoneware as well as for greenhouse workers who commercialize seedlings of the species. Concerning substrates under investigation, the one that had the best result, with the highest frequency of significant interactions with IBA in the variables, was carbonized rice husk with vermicompost and sand (CVS) with three interactions, followed by carbonized rice husk with vermicompost (CV) and vermiculite (V), which had only one interaction (Figure 1). The rooting variable was the one that presented the most visible interactions in the results between the evaluated variables. The probable cause of this effect appearing in the rooting rate and not in the other variables may be related to the fact that the IBA directly interferes only in the rooting and not in the other parameters observed (HARTMANN et al., 2011). A significant interaction was found when IBA was combined with V, CVS, and CV (Figure 1A). Regarding the use of the IBA, Holanda et al. (2012) found equivalent results since IBA influenced the rooting and survival of Brazilian peppertree seedlings positively. However, 90 days after planting, a part of the cuttings have calluses; it indicated that roots could be forming and that results could be better than the ones found by Holanda et al. (2012), who evaluated cuttings 100 days after planting.

Table 1. Generalized Linear Model Results for the presence or absence of IBA in Brazilian peppertree cuttings.

	Estimate	Standard Error	Z value	Pr(>|z|)
Rooting percentage				
	Estimate	Standard Error	Z value	Pr(>|z|)
(Intercept)	1.94	0,23	-8,14	3,94x10-16*
IBA	1,44	0,29	4,96	7,15x10-7*
(Dispersion parameter for binomial family taken to be 1)				
AIC = 336,27				
Buds presence				
(Intercept)	1,87	0,38	4,93	8,25x10-2
IBA	1,02	0,62	1,66	0,10
(Dispersion parameter for binomial family taken to be 1)				
AIC = 75,55				
Number of buds				
(Intercept)	-0,43	0,27	-1,55	0,12
IBA	1,00	0,29	3,40	6,68x10-4*
(Dispersion parameter for Poisson family taken to be 1)				
AIC = 243,61				
Number of leaves				
(Intercept)	1,31	0,12	10,75	2,00x10-16*
IBA	0,65	0,13	4,96	6,72x10-7*
(Dispersion parameter for Poisson family taken to be 1)				
AIC= 521,96				
Length of the largest root (cm)				
	Estimate	Standard Error	t value	Pr(>|t|)
(Intercept)	4,03	1,53	2,62	0,01*
IBA	5,95	1,77	3,35	1x10-3*
(Dispersion parameter for gaussian family taken to be 47.22)				
AIC = 539,40				
Dry weight of roots (g)				
(Intercept)	0,01	0,02	0,71	0,48
IBA	0,11	0,02	3,87	5,94x10-4*
(Dispersion parameter for gaussian family taken to be 0.02)				
AIC = -64,41				

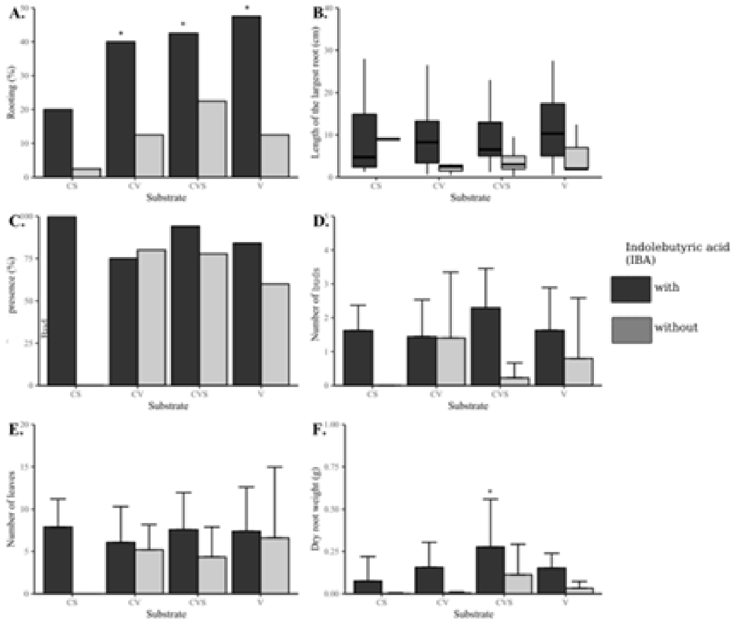
(*) A statistically significant difference (GLM; P<0.05).

It has been known that callus development (callogenesis) is the first visible morphological alteration in cuttings in the process of cell differentiation and formation of radicular emergences (rhizogenesis) (GOULART et al., 2014). Despite that, the evaluation that should be conducted 90 days after planting was pre-established because, from the commercial perspective, delay in rooting is a negative factor in the production of seedlings by cutting.

Results that differ from those found by Holanda et al. (2012) can be caused by the size of the stake (FACHINELLO et al., 1995). Because thicker woody cuttings may have contributed to the success of rooting, as they may have high levels of resources and nutrients.

Concerning the length of the longest root (cm), bud percentage (%), and the number of leaves (length ≥ 1 cm), statistically significant interactions were not observed among factors (Figure 1B, 1C, and 1E). However, all cuttings with IBA and commercial substrate (CS) yielded buds, but the number of buds per cutting was smaller than the one of IBA and CVS, the only one that got significant interaction among all

Figure 1: (A.) Rooting percentage in Brazilian peppertree cuttings. (B.) Length of the largest root (cm) in Brazilian peppertree cuttings. (C.) Percentage of bud presence in Brazilian peppertree cuttings. (D.) Number of buds per Brazilian peppertree cutting. (E.) Number of leaves per Brazilian peppertree cutting. (F.) Dry weight of roots (g) of Brazilian peppertree cuttings. Legend: Vermiculite (V); Carbonized rice husk with bovine vermicompost (CV); Carbonized rice husk with vermicompost and sand (CVS); Commercial substrate (CS); Significant interaction P(Z)<0.05 or P(t)<0.05 (*).



combinations (Figure 1D). Another variable that had a statistically significant interaction between IBA and CVS was root dry mass (g) (Figure 1F).

For the variables evaluated, the high range found in the data set may be the result of the use of cuttings from different matrices, a fact that generates variability among treatments. According to Fanchinello et al. (2005), factors that may influence the rooting of plant cuttings include genotype, age, and health, besides environmental and physiological conditions of the matrix.

The substrate is a fundamental factor to improve the rhizogenesis process and adequate radicular system development. In this study, the substrate that had the best result was carbonized rice husk with vermicompost and sand (CVS). Rice husk has adequate contents of potassium and calcium, two macronutrients that are essential for plant development, along with its highly alkaline pH, factors that make it beneficial in the substrate composition (SAIDELLES et al., 2009). On the other hand, sand is usually inert or a little fertile, with low density and high porosity. It is commonly used (pure or in the composition of mixed substrates) in rooting plant cuttings, thus contributing to aeration and infiltration (MARANHO; PAIVA, 2011). Vermicompost, which is obtained through the assimilation of bovine waste by worms, provides important nutrients, such as phosphorus, calcium, magnesium, potassium, zinc, and copper, besides humic acids, to plants (FRANCESCHI et al., 2018). It improves the chemical and physical characteristics of substrates and is also responsible for decreasing apparent density and increasing porosity and water retention (JOSHI et al., 2015).

According to Franceschi et al. (2018), the use of bovine vermicompost in the ratios of 50% and 75% with commercial substrate favored the growth of seedlings resulting from Brazilian peppertree seeds. The authors have attributed the increase in growth to the high amounts of nutrients found in the vermicompost. Studies have shown the benefit of the use of vermicompost in the composition and fertilization of substrates in the production (either sexual or asexual) of tree species (ANTUNES et al., 2016, AMARAL et al., 2017).

The commercial substrate (CS) was the one that exhibited the lowest development. Even though this substrate – composed of peat and carbonized rice husk – has good composition and nutrient levels , however, it showed a low rooting rate . Verdonck et al. (1984) have referred to the decrease in quality of substrates with high peat content, regarding root porosity and aeration. Lima et al. (2016) found low results of rooting of *Langerstroemia indica* L. cuttings in an organic commercial substrate with peat, by comparison with vermiculite. It should be highlighted that, according to the manufacturer, this substrate aims at germination and production of vegetable seedlings, rather than rooting of plant cuttings, even though Brandão et al. (2020) have satisfactory results of commercial substrates with peat in the composition they used to root *Malpighia emarginata* DC. cuttings.

Results show that the use of IBA along with porose substrates that have a high content of organic matter and nutrients, such as vermicompost, favored the rooting of Brazilian peppertree cuttings. However, novel studies of different substrates (pure and combined ones) and other dosages of IBA should be conducted in the search for better results in rooting and early development of seedlings. Other factors, which were not considered in this study, should be evaluated, since they may influence cutting development.

CONCLUSION

1. The use of IBA in rooting of Brazilian peppertree cuttings is recommended since it was beneficial to most variables evaluated in all substrates under study.

2. The substrate that got the best interaction with IBA, among the variables, was carbonized rice husk with vermicompost and sand; thus, its use is recommended in cutting. Based on the results, the use of a commercial substrate with peat is not recommended.

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