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Propagation of pariparoba (Piperaceae) by different types of stem cuttings and substrates

Propagação de pariparoba (Piperaceae) por diferentes tipos de estacas caulinares e substratos

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ABSTRACT
Piper cernuum Vell., a native species known as *pariparoba*, presents essential oil with potential properties and significant growth in clearings, being indicated for the recovery of degraded areas. Prospecting studies for new botanical matrices need the development of specific essays of plant propagation for future domestication of the species. This work aimed to evaluate the rooting of different types of cuttings using different substrates to develop a seedling production protocol. Stem cuttings with one or two nodes were installed on three types of substrates (medium sifted sand, vermiculite of medium granulometry and commercial substrate [Tropstrato HP®]) and maintained in greenhouse for 60 days to evaluation of the percentages of survival, mortality, sprouts, rooting, average number and average length of the roots. The results showed interaction between cutting types and substrates for rooting percentage and sprouting cuttings. The largest rooting percentages were observed in apical cuttings planted in sand and vermiculite (57.0 and 55.25%, respectively). Cuttings with two nodes presented higher performance than those with one node when compared for the analyzed variables.
Keywords: cutting; native pepper; nodes; non-timber forest products (NTFPs).

RESUMO
Piper cernuum Vell., espécie nativa conhecida como pariparoba, possui óleo essencial com propriedades potenciais e crescimento significativo em clareiras, sendo indicada para recuperação de áreas degradadas. Estudos de prospecção para novas matrizes botânicas necessitam de ensaios específicos de propagação vegetativa para futura domesticação da espécie. Esta pesquisa teve como objetivo avaliar o enraizamento de diferentes tipos de estacas e substratos para desenvolvimento de protocolo de produção de mudas. Instalaram-se estacas caulinares, com um ou dois nós, em três tipos de substratos (areia média peneirada, vermiculita de granulometria média e substrato comercial [Tropstrato HP®]). As estacas foram mantidas em casa de vegetação por 60 dias para avaliação da porcentagem de sobrevivência, mortalidade, brotações, enraizamento, número médio e comprimento médio de raízes. Os resultados mostraram interação entre tipos de estacas e substratos para estacas enraizadas e estacas com brotações. As maiores porcentagens de enraizamento foram obtidas nas estacas apicais em areia e vermiculita (57,0 e 55,25%, respectivamente). Estacas com dois nós apresentaram rendimentos superiores quando comparadas àquelas com um nó para as variáveis analisadas.
Palabras-chave: estaquia; nós; pimenta nativa; produtos florestais não madeiráveis (PFNMs).

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INTRODUCTION

Prospecting plant resources from native flora can significantly increase the phytochemical potential from a social and socio-environmental perspective for communities living in conservation areas (RAUT & KARUPPAYIL, 2014). For this purpose, studies are required regarding the chemical composition and the cultivation of the species, in order to carry out their efficient and sustainable management.

Piper genus is used all over the world, with wide distribution in the Americas and Asia. The composition of the oils of the different species in Brazil has been researched under several biological aspects (MESQUITA *et al*., 2005; PEREIRA *et al*., 2008; GUERRINI *et al*., 2009; OLIVEIRA *et al*., 2013; GIROLA *et al*., 2015).

The species *Piper cernuum* Vell., popularly known as *pariparoba,* is a native species not endemic in Brazil (FLORA DO BRASIL 2020 EM CONSTRUÇÃO, 2018). It has a growth habit that can reach 6 meters, has leaves up to 40 cm in length, is found in primary and secondary forests and its traditional use includes analgesic action for stomach, liver, kidney and circulatory problems (MARIOTT *et al*., 2003).

Studies of its population dynamics indicate that its growth is dependent on high luminosity, a condition usually found in clearings, which points to its use for the recovery of degraded ecosystems (MARIOTT *et al*., 2007).

Anatomical analysis of *Piper cernuum* was performed by Gogosz *et al.* (2012) who characterized the species by the presence of sinuous epidermis with thin cuticle, long trichomes, absence of starch and presence of phenolic compounds, that are peculiar characters for its correct identification.

Bizzo *et al*. (2009) pointed out that essential oils have been researched and used in several segments for only a small number of species, compared to the high diversity of aromatic flora in Brazil, making thus relevant the development of technology for future production of these compounds.

Comparing species of the *Piper* genus, Mesquita *et al*. (2005) identified 32 compounds in the essential oil of *Piper cernuum*, with the constituents trans-dihydroagarofuran (22.4%), 10-epi-γ-eudesmol (16.8%), germacrene D (9%), Elemol (7.2%) and alpha-pinene and β-eudesmol (4.1% each), whose properties indicate antimicrobial, insecticide and anti-inflammatory activities respectively.

Pharmacological studies conducted by Girola *et al*. (2015) described the constituent camphene present in this species as responsible for the induction of apoptosis in tumor cells, indicating a new perspective for anticancer therapy.

In this context, the prospection and rational use of plant resources requires vegetative propagation experiments that may favor the production of seedlings from specific protocols, since adventitious rooting is dependent on physiological and environmental factors (ZUFFELLATO-RIBAS & RODRIGUES, 2001).

Piper species have different forms of propagation and there are few specific studies about this subject, as pointed by Souza *et al.* (2009). So this research aimed to evaluate the rooting of cuttings of *Piper cernuum* with different lengths and installed in different substrates.

MATERIAL AND METHODS

The collection of branches with leaves from mature plants of *Piper cernuum* Vell. (MBM396416 register) was carried out in February 2016, at Bom Jesus Biological Reserve, Antonina, State of Paraná, Brazil. The plant material was moistened and conditioned in black polyethylene bags to be transported to the greenhouse in the Agrarian Sciences sector of the Federal University of Paraná, where it was kept under intermittent mist until the propagules were prepared.

Three types of stem cuttings were used, corresponding to the treatments: semihardwood stem cuttings of the median region (without leaves and containing 1 or 2 nodes), and apical herbaceous cuttings (without nodes and containing a leaf reduced to a quarter of its original area). The cutting of the cuttings was standardized diagonally (bevel) at the base and rectum in the upper portion.

Cuttings with one node were made with a length of 5 to 7 cm whereas those of 2 nodes varied from 10 to 12 cm. Apical cuttings were standardized as being at 7 cm of the base until the insertion

of the canaliculate petiole. The diameter of the cutting was 2.0 ± 0.3 cm and the apical diameter was 1.0 ± 0.3 cm. After cutting, the propagules were washed in running water for five minutes. Solutions containing plant regulators were not used due to the intention to produce of seedlings in an Environmental Protection Areas (EPA), where contaminant inputs are prohibited.

Planting was carried out in plastic tubes of 120 cm^3 volume, filled with the three evaluated substrates: medium sifted sand, vermiculite of medium granulometry and commercial substrate (Tropstrato HP®). The tubes were arranged alternately on plastic supports with a capacity of 96 tubes. The cuttings were kept in greenhouse with intermittent nebulization of 5 seconds every 30 minutes.

The experimental design was completely randomized in a 3x3 factorial arrangement (three types of cuttings and three substrates), with four replications and 14 cuttings per plot, comprising 56 cuttings per treatment, and a total of 506 cuttings in the experiment.

After 60 days, the percentages of survival, mortality and sprouts, average number and average length of the three largest roots were evaluated. Data were submitted to analysis of variance homogeneity by the Bartlett test, variance analysis with Anova and mean comparison by the Tukey test at 5% probability. The statistical analysis were carried out with Assistat (SILVA & AZEVEDO, 2016).

RESULTS AND DISCUSSION

The interaction between substrates and cuttings types was significant for the variables rooting percentage and cuttings with sprouts. Cuttings types influenced all variables analyzed while the substrate types influenced the rooting percentage and the number and length of roots (table 1).

Table 1 – Rooting percentages, mortality, sprouting, number of roots and average length of the three largest roots in different cuttings of *Piper cernuum* as a function of three substrates.

Substrates	Cuttings type			Mean
	1 node	2 nodes	Apical	
		Rooting (%)		
Sand	8.80 aC	28.50 aB	57.00 aA	31.43
Tropstrato	8.75 aC	30.00 aB	40.75 bA	26.50
Vermiculite	14.00 aC	28.25 aB	55.25 aA	32.50
Mean	10.52	28.92	51.00	
		Mortality (%)		
Sand	91.20	71.50	43.00	68.23^{ns}
Tropstrato	91.25	70.00	59.25	73.40
Vermiculite	86.00	71.75	44.75	89.50
Mean	89.48 a	71.08 b	49 c	
		Sprout cuttings (%)		
Sand	5.30 aB	19.58 aA	26.50 aA	17.13
Tropstrato	8.75 aA	19.25 aA	7.00 bA	11.67
Vermiculite	10.50 aA	21.00 aA	14.00 abA	15.17
Mean	8.18	19.94	15.83	
		Roots number (n)		
Sand	6.00	9.78	9.93	8.57 ab
Tropstrato	4.50	7.05	7.96	6.50 b
Vermiculite	6.95	8.73	12.68	9.45 a
Mean	5.82 b	8.52 a	10.18 a	
		Roots lenght (cm)		
Sand	2.48	5.38	6.13	4.66 a
Tropstrato	2.20	3.18	4.70	3.36 b
Vermiculite	2.63	5.83	7.33	5.26 a
Mean	2.43 b	4.79 a	6.05 a	

* Means followed by the same uppercase letter in the rows and lowercase in the columns do not differ from each other by the Tukey test at 5% probability. Ns: not significant.

Larger rooting percentages were observed in apical cuttings when planted in washed sand and vermiculite (57.0 and 55.25%, respectively). Lower values were observed for cuttings with one node in all evaluated substrates. Higher mortality rates were also observed in cuttings with one node, followed by cuttings with two nodes, and apical cuttings showed the highest survival rates (51%).

In the sand substrate, cuttings with 1 node had a lower percentage of sprouts compared to 2-node cuttings and apical cuttings. Cuttings with 2 nodes showed lower variations for this variable, with a value close to 20%. Apical cuttings had their budding ability reduced (7%) when planted on Tropstrato substrate. Lower mean number and root length were also observed in the commercial substrate. Regarding the types of cuttings, 2-node cuttings and apical cuttings showed a greater number and length of roots compared to 1-node cuttings (table 1).

The sand and vermiculite substrates presented values higher than 50% for the variable rooting, agreeing with the results obtained by Cunha *et al*. (2015) for the species of the same genus, *Piper hispidum*. These results demonstrate that the rooting of some species depends on materials that have higher porosity and consequent aeration of the lower portion of the pile, a condition pointed out by Kämpf (2000) in the comparison between different types of substrates.

Apical cuttings of *P. cernuum* presented the highest percentage of rooting when compared to cuttings with two nodes and with one node, which obtained the smallest percentage (figure 1). These juvenile propagules were the only type that presented foliar bud pores in their structure, pointed as promoters of cofactors synergistic synthesis of the rooting induction process. As mentioned by Taiz & Zeiger (2013), the greater juvenility of the apical material occurs due to the synthesis of auxins in the meristematic regions that facilitate the physiological status of the propagule, favoring its rooting. The same condition was observed by Bischoff *et al.* (2017) on cuttings of *Varronia curassavica* Jacq. (Boraginaceae), where the best performance of the propagules was obtained from apical buds. According to Hartmann *et al*. (2011), the presence of leaves in the apical material constitutes a source of carbohydrates and cofactors for the rhizogenic process.

Figure 1 – *Piper cernuum* rooted cuttings: (A) apical; (B) one node; (C) two nodes.

Within the *genus Piper*, different responses regarding the type of cuttings and substrates used were verified. Thus, Cunha *et al*. (2015) obtained results superior to 80% with the use of apical and basal cuttings in sand and vermiculite and Gomes & Krinski (2016) reported for *Piper amalago* L. maximum percentages of 22.92% in the interaction between basal cuttings and soil substrate, being vermiculite the worst performing substrate, and for *Piper umbellatum* L., values of up to 53.8% using 15 cm stem cuttings. These results justify the exploratory experiments for each species.

Considering cuttings with two nodes of *P. cernuum* and the branch thickness, it is possible to infer that cuttings with longer lengths kept the reserves required for the period demanded for the formation of adventitious roots, to the detriment of those with only one node. These results differed partially from the results obtained for the species *Potomorphe umbellata* (Piperaceae), where cuttings with one node did not present a slight superiority in relation to those of two nodes (MATTANA *et al*., 2009).

It should be noted that although the use of apical cuttings unveiled the best results, it is necessary to take into account the optimization of the use of all the branches for a maximum yield, especially when one evaluates the preservation of the species and the saving of vegetal material for propagation by cutting, agreeing with the statement of Mattana *et al*. (2009).

CONCLUSION

For the production of *Piper cernuum* seedlings, apical cuttings installed in sand or vermiculite substrates are indicated. Cuttings with two nodes presented higher performance than those with one node for the analyzed variables.

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