

SCIENTIFIC NOTE

Effect of cuttings defoliation and different substrates on the vegetative propagation of the monkey-pepper (*Piper aduncum* L.) (Piperaceae)

Efecto de la defoliación de esquejes y diferentes sustratos en la propagación vegetativa de pimienta de mono (*Piper aduncum* L.) (Piperaceae)



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**Cuttings type and rooting performance on
Piper aduncum L.**

Photo: A.P. Ferriani

ABSTRACT

Forests constitute a valuable natural resource, especially when including non-timber forest products (NTFPs). Bioprospecting for and sustainable exploration of native species and the development of protocols for seedling production promote conservation. *Piper aduncum* L. stands out in several regions of Brazil because of several biological activities, notably action as a repellent, antimicrobial and insecticide. This study aimed to evaluate the influence of defoliation and different substrates on the cutting process. For this, an experiment was conducted in February, 2016 at the Health Plant Department, Sector of Agricultural Sciences, Federal University of Paraná (UFPR). *Piper aduncum* stem cuttings with 10 cm and 1/3 of the leaf area and defoliated cuttings. Stem cuttings were washed in running water for 5 minutes for subsequent use in three different substrates (medium sand sifted, vermiculite and commercial substrate Tropstrato HP®). The experiment design was completely randomized in a 2×3 factorial arrangement (cutting type x substrate), and it was evaluated the rooting percentage, mortality, shoot emission, number of roots and average length of the three largest roots after 60 days. The results confirmed that

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the presence of leaves in *P. aduncum* stem cuttings promotes adventitious rooting. The different substrates did not exert a significant influence on the performance of the cuttings in this species.

Additional key words: propagation by cuttings; pruning; growing media; non-timber forest products (NTFPs); native spice.

RESUMEN

Los bosques constituyen un valioso recurso natural, principalmente cuando son productos forestales no maderables. La bioprospección y exploración sostenible de especies nativas, pueden desarrollar protocolos de producción de plántulas al potenciar la conservación de áreas de interés. La especie *Piper aduncum* L. se encuentra en varias regiones, presenta diversas actividades biológicas, al destacarse acciones repelente, antimicrobiana e insecticida. El objetivo de este trabajo fue evaluar la influencia de la defoliación y diferentes sustratos en el proceso de propagación. Para esto, un experimento fue realizado en febrero de 2016 en el Departamento de Fitotecnia y Fitosanidad, Sector de Ciencias Agrarias, Universidad Federal de Paraná. Se utilizaron esquejes semileñosas de *Piper aduncum* con 10 cm y un 1/3 del área foliar y defoliación total. Los esquejes fueron lavados por 5 minutos en corriente de agua para posteriormente usados en tres diferentes sustratos (arena tamizada, vermiculita y sustrato comercial Tropstrato HP®). El diseño experimental fue completamente al azar con arreglo factorial 2x3 (tipo de esqueje x sustrato), y fue evaluado el porcentaje de enraizamiento, mortalidad, emisión de brotes, número de raíces y longitud media de las tres raíces más largas después de 60 días. Los resultados indicaron la necesidad de las hojas en los esquejes de *P. aduncum* para promoción del enraizamiento adventicio. Los sustratos arena, vermiculita y Topstrato no presentaron influencia significativa en el desempeño de los esquejes de la especie.

Palabras clave adicionales: propagación por estacas; poda; sustratos de cultivo; productos forestales no maderables (PFNM); pimienta nativa.

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INTRODUCTION

Forests, in general, constitute one of the most valuable natural resources. In addition to being biodiversity centers, they are important climate regulators, rainwater drainage, and efficient soil protectors. They are also essential sources of resources for man, producing, for example, wood, cellulose, resins, tannins and essential oils, among other non-timber forest products (NTFP) (Backes, 2009).

Prospecting native species with potential for use as NTFP seeks to minimize the impact of predatory extractivism of natural areas and subsidize the exploration of new compounds of interest in a sustainable manner. The species with potential for production of compounds of interest include the genus *Piper* (Piperaceae), which occupies a prominent position. Fazolin *et al.* (2006) highlighted the variety of compounds

from mixed routes (chiquimate/acetate) generating phenylpropanoic aromatic compounds (lignans and neolignans), as well as terpenes and flavonoids. For this reason, the composition of essential oils of different species of Piperaceae in Brazil has been researched under several biological and biotechnological aspects (Mesquita *et al.*, 2005; Pereira *et al.*, 2008; Guerrini *et al.*, 2009; Oliveira *et al.*, 2013; Girola *et al.*, 2015; Krinski *et al.*, 2018a).

Piper aduncum L., popularly known as Monkey Pepper, is native to Tropical America and widely distributed throughout the Brazilian territory. It is considered an ombrophilous species, occurring in several forest formations, preferably in soils with a high organic matter content and humidity (Lorenzi and Mattos, 2002). Morphologically, this species is described as a

bush or small tree, from 2 to 7 m, showing several nodes, with membranous or chartaceous, elliptic or elliptic-oval leaves (Maia *et al.*, 2000).

This species presents great potential in the recomposition of degraded areas, being a colonizer of altered areas that promotes greater natural regeneration and relative density over time (Alvarenga *et al.*, 2006). Nevertheless, the main economic interest of this species is due to the production of essential oil with varied biological properties. There are reports in the literature of yields of up to 3.5% of essential oil, and the phenyl ether dillapiol is the most frequently reported major constituent (Sousa *et al.*, 2008; Almeida *et al.*, 2009). Among the numerous biological activities attributed to *P. aduncum* oil, repellent actions (Misni *et al.*, 2009), antimicrobial agents (Guerrini *et al.*, 2009) and insecticide actions stand out (Pereira *et al.*, 2008; Mesquita *et al.*, 2005; Krinski and Foerster, 2016; Sanini *et al.*, 2017; Krinski *et al.*, 2018b).

According to Elias and Santos (2016), for NTFPs to present a viable alternative that stimulates conservation and promotion of community development, some relevant aspects need to be clarified, including botanical, ecological and agronomic or silvicultural knowledge of this species. In this sense, knowledge on appropriate propagation techniques is important because it is one of the initial stages in the process of domestication of species of interest, aiming at a more rational exploitation that is not based only on extractivism.

In order to provide specific knowledge on species propagation and phytochemical research, the objective of the present study was to evaluate the influence of the presence of leaves or defoliation and the use of different substrates on the vegetative propagation of *P. aduncum* using stem cutting.

MATERIAL AND METHODS

The collection of branches with leaves from adult *Piper aduncum* plants (MBM396411 register) was carried out in February, 2016, at the Bom Jesus Biological Reserve, Antonina, Parana, Brazil. The plant material was moistened and conditioned in black polyethylene bags for transport to a greenhouse in the Agrarian Sciences sector of the Federal University of Parana, where it was kept under intermittent misting until the propagules were prepared.

Semiwoody stems generated cuttings with a 10 cm length and standard diagonal cut (bevel) at the base, straight in the upper portion, with 1/3 of the leaf area or total defoliation. After cutting, the propagules were washed in running water for 5 min. Solutions containing plant regulators were not used because the seedlings were produced in Environmental Protection Areas (EPA) where contaminant inputs are prohibited.

The planting was carried out in plastic tubes, 120 cm³ volume, with the three substrates: medium sand sifted, vermiculite and commercial substrate Tropstrato HP®. The cuttings were kept in a greenhouse with an intermittent nebulization of 5 s every 30 min.

The experiment design was completely randomized in a 2×3 factorial arrangement (presence or absence of leaves and three substrates), with four replications and 14 cuttings per plot, comprising 56 cuttings per treatment and a total of 336 cuttings in the experiment.

After 60 d, the percentages of survival, mortality and sprouts, average number and average length of the three largest roots were evaluated. The data were submitted to analysis of variance homogeneity with the Bartlett test, variance analysis with ANOVA and means comparison with the Tukey test at 5% probability. The statistical analysis was carried out with Assistat (Silva and Azevedo, 2016).

RESULTS AND DISCUSSION

The analysis of variance showed that there were no differences among the substrates used; however, the presence of leaves was significant for all variables, except for percentage of cuttings with shoots. The interaction between the factors (presence or absence of leaves and substrates) was significant only for cutting mortality (Tab. 1).

The rooting percentages in the *P. aduncum* cuttings ranged from 2.4 to 30.8%. The highest averages were recorded in cuttings with the presence of leaves (Tab. 2) and confirmed the interespecific variation in *Piper* plant propagation, as pointed out by Dosseau (2009), Chaves *et al.* (2014), Cunha *et al.* (2015), Gomes and Krinski (2016a, 2016b), and Ferriani *et al.* (2019).

Ferriani *et al.* (2018) also verified an increase in rooting percentage responses in cuttings with leaves in

Table 1. Analysis of variance for rooting percentage (ROO), percentage of mortality (MOR), percentage of cuttings with shoot emission (SHO), number of roots (NUM) and length of the three largest roots (LEN), on cuttings of *Piper aduncum* with and without leaves depending on different substrates. Curitiba-PR (2016).

Source of variation	DF	F values				
		ROO	MOR	SHO	NUM	LEN
Presence of leaves (L)	1	26,45**	25,79**	0,17 ^{NS}	34,17**	55,22**
Substrates (S)	2	1,40 ^{NS}	0,50 ^{NS}	0,17 ^{NS}	2,84 ^{NS}	1,47 ^{NS}
Interaction (LxS)	2	1,40 ^{NS}	4,79*	1,21 ^{NS}	0,31 ^{NS}	2,02 ^{NS}
Residue	12	-	-	-	-	-
CV (%)		54,21	7,52	145,34	40,79	44,30

** significant at 1%; * significant at 5%; ^{NS}: not significant. DF: degrees of freedom; CV: coefficient of variation.

Table 2. Mean and standard deviation of rooting percentage, mortality, sprout emission, number of roots and mean length of the three largest roots on cuttings of *Piper aduncum* with and without leaves depending on different substrates. Curitiba-PR (2016).

Cuttings	Substrates			Mean
	Sand	Vermiculite	Topstrato	
Rooting (%)				
With leaf	30,8±4,1 aA	18,9±4,1 aA	18,9±14,8 aA	22,9±2,9 a
Without leaf	4,8±4,1 bA	7,1±7,1 aA	2,4±4,1 bA	4,7±2,2 b
Mean	17,8±4,1 A	13,0±4,5 A	10,7±6,4 A	
Mortality (%)				
With leaf	71,4±7,1 bA	81,0±4,1 aA	76,2±10,9 bA	76,2±8,0 b
Without leaf	95,2±4,1 aA	83,3±4,1 aA	95,2±4,1 aA	91,3±6,9 a
Mean	83,3±14,0 A	82,1±3,9 A	85,7±12,8 A	
Sprout cuttings (%)				
With leaf	7,1±12,3 aA	2,4±4,1 aA	4,7±8,2 aA	4,7±7,9 a
Without leaf	2,4±4,1 aA	11,8±10,8 aA	4,7±4,1 aA	6,3±7,5 a
Mean	4,7±8,6 A	7,1±8,9 A	4,7±5,8 A	
Roots number (n)				
With leaf	5,5±0,5 aA	7,1±1,5 aA	5,2±1,1 aA	5,9±1,3 a
Without leaf	2,0±2,0 bA	2,7±2,5 bA	0,3±0,6 bA	1,7±1,9 b
Mean	3,8±2,3 A	4,9±3,1 A	2,8±2,8 A	
Roots length (cm)				
With leaf	7,7±1,4 aA	9,3±3,7 aA	11,3±2,7 aA	9,4±2,9 a
Without leaf	0,3±0,6 bA	3,2±3,2 bA	0,03±0,06 bA	1,2±2,2 b
Mean	4,0±4,1 A	6,2±4,1 A	5,7±6,4 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 ($P \leq 0.05$).

three species of *Piper* (*P. arboreum*, *P. cernuum* and *P. diospyrifolium*) with indolbutyric acid (IBA) use, highlighting new study perspectives.

The mortality of *P. aduncum* cuttings was also lower in cuttings with the presence of leaves on the substrates sand and Topstrato, with no difference in the

cuttings with and without leaves in the vermiculite substrate; however, a significant difference was observed for this variable in the commercial substrate (Tab. 2).

The percentage of cuttings with shoots did not present significant difference for this species because

there were percentages of rooting or mortality that were significant and complementary (Tab. 2). In addition to higher rooting percentages, the presence of leaves in the *P. aduncum* cuttings promoted increases in the number and mean of the root length (Tab. 2).

The values of the rooting percentage observed for *P. aduncum* in our study, when compared with other species of the genus, were relatively low; however, the literature shows a high variation in the rooting potential of this species.

Stem cuttings of *Piper hispidum* Sw. have shown percentages higher than 85% (Cunha *et al.*, 2015). Pescador *et al.* (2007) reported values higher than 60% for *Piper mikianium* (Kunth). Steudel. Chaves *et al.* (2014) evaluated stem cuttings of three species of the genus for rhizogenic potential, where *Piper hispidum* Sw. and *Piper tuberculatum* Jacq. presented values higher than 80%, while *Piper marginatum* Jacq. did not exceed 20% rooting.

With values similar to those observed in the present study, Mattana *et al.* (2009) reported rooting values of up to 37.5% for *Piper umbellatum* L. However, Gomes and Krinski (2016a), in the same species, obtained values between 37.5 and 60% rooting, without influence from the substrates. The longest cutting length (20 cm) had the best performance.

Even so, for *P. aduncum*, the best rooting performance in the leaf cuttings occurred because these organs are sites of synthesis of auxin and carbohydrates translocated to the base of the cuttings, accelerating the process of rooting and reducing the mortality rates (Hartmann *et al.*, 2010). In addition to the synthesis of auxin and carbohydrates, it is probable that rooting and greater vigor in leaf cuttings are related to the synthesis of phenolic compounds that interact with auxin, inducing a greater vigor and percentage of root emission (Pacheco and Franco, 2008).

The significant difference for the mortality of the cuttings in the commercial substrate corroborated with the results obtained by Purcino *et al.* (2012) for two species of the genus *Ocimum* (*O. gratissimum* and *O. selloi*). Overall, the survival of the cuttings was low, differing from the values found by Dosseau (2009), who reported up to 90% survival in medium plagiotropic cuttings for *P. aduncum* in a sand substrate. Nonetheless, the author reported up to 100% mortality for basal cuttings in a vermiculite substrate.

The increase in the number of roots and mean root length verified in our study is in agreement with other studies, such as the one by Vignolo *et al.* (2014), who reported a greater number and length of roots in cuttings of three mulberry cultivars with the presence of leaves, to the detriment of cuttings without leaves.

When analyzing the percentage of cuttings with shoots, although there were no significant differences for *P. aduncum*, other species, such as those of the genus *Ocimum* studied by Purcino *et al.* (2012), have had cuttings with whole leaves or leaves reduced by half that presented percentages of shoots higher than 97 and 1.3% live cuttings, while seedlings without leaves have maintained rates of 65 and 30%. These results corroborated the relationship between leaf maintenance, as a prerequisite for sprouting, and subsequent rooting.

Together with this factor, the longest and number of roots are probably related to a higher endogenous auxin concentration because of the presence of leaves, which favors the best development of the root system (Antunes *et al.*, 2000).

In addition, in *P. aduncum*, there was greater rooting in the sand substrate and higher values for the number and length of roots in the vermiculite substrate. In another *P. aduncum* study in the length of propagules and substrates, Ferriani *et al.* (2019) observed higher rooting responses (26.5%), a lower length (10 cm), without influence from the substrates and lower results with the longest cuttings (20 cm) in a vermiculite substrate.

These data are a bit different those reported by Dosseau (2009), who found higher values of survival and number and length of roots in orthotropic and plagiotropic stem cuttings of *P. aduncum* in a sand substrate, as compared to vermiculite. However, in both cases, it was observed that substrates with higher porosity presented satisfactory results.

These discrepancies in the rooting percentages showed the different genetic potentials of each species for adventitious root emission and suggested the need for specific studies for each species in order to define adequate propagation protocols (Gomes and Krinski, 2016a; 2016b; 2018a; 2018b; 2019). Further studies are indicated because of the biological and environmental importance reported in many native *Piper* species.

Thus, because of the scarcity of studies on the vegetative propagation of *P. aduncum*, complementary studies involving rejuvenation techniques, application of plant regulators and the dynamics of seasonality should be carried out on the rooting of this species.

CONCLUSIONS

The presence of leaves in *P. aduncum* stems promotes adventitious rooting. The substrates sand, vermiculite and Topstrato did not present a significant influence on the performance of *P. aduncum* cuttings under the conditions in which this experiment was carried out.

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Conflict of interests: the manuscript was prepared and reviewed with the participation of the authors, who declare that there exists no conflict of interest that puts in risk the validity of the presented results.

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