Does the presence of leaves on coleus stem cuttings influence their rooting? (1)

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ABSTRACT

Plectranthus scutellarioides (L.) R. Br. is part of the Lamiaceae family and is commonly known as coleus, due to the genus denomination in the past. This ornamental plant species has been gaining importance in the floristic industry due to the great number of exotic cultivars available in the market. Coleus commercial exploitation by seeds is held up by genetic instability, which makes it usually being propagated through semi-hardwood stem cuttings and leaves have great importance in stem cuttings adventitious rooting. The objective of this study was to evaluate the presence or absence of apical leaves and the use of indolebutyric acid (IBA) in coleus stem cuttings rooting. Semi-hardwood coleus stem cuttings with 6 cm long were prepared with or without apical leaves, and then treated with hydroalcoholic solutions (50% v v^{-1}) of 0, 1000 and 2,000 mg L^{-1} IBA. The propagules were planted in tubes containing vermiculite and kept for 30 days in a greenhouse under intermittent misting (24 ± 2 °C, Relative Humidity 90%) until final evaluation. The experiment was conducted in a completely randomized design, in a 3x2 factorial scheme (3 IBA concentrations x presence and absence of apical leaves). The rooting was evaluated after 30 days. Stem cuttings with and without apical leaves has 100 and 57% rooting, respectively. Stem cuttings rooting did not vary according to IBA concentrations. Roots length and number were higher in stem cuttings with apical leaves in comparison to leafless ones, regardless IBA concentrations. On average, 97.7% sprouting was observed in cuttings with apical leaves, significantly higher rates when compared to the average of 2.2% on leafless cuttings. The presence of apical leaves is fundamental for coleus stem cuttings rooting and the use of IBA is not required for this species propagation.

Keywords: *Plectranthus scutellarioides*, auxin, ornamental, rhizogenesis, vegetative propagation.

RESUMO

A presença de folhas apicais nas estacas de coleus influencia o enraizamento?

Plectranthus scutellarioides (L.) R. Br. pertence à família Lamiaceae e é comumente conhecida como coleus, devido à antiga denominação do gênero. Essa planta ornamental vem ganhando destaque na indústria florística em função do grande número de cultivares exóticas no mercado. Sua exploração comercial por meio de sementes é dificultada pela instabilidade genética, sendo propagada vegetativamente com estacas semilenhosas e as folhas têm grande importância no enraizamento de estacas. O objetivo deste trabalho foi avaliar a presença e ausência de folhas apicais e do uso de ácido indol butírico (IBA) no enraizamento de estacas caulinares de coleus. Estacas semilenhosas de coleus com 6 cm de comprimento, com e sem de folhas apicais, tratadas em solução hidroalcoólica (50%) de 0, 1000 e 2000 mg L^{-1} de IBA foram confeccionadas. Os propágulos foram estaqueados em tubetes com vermiculita e mantidos em casa de vegetação (24 ± 2 °C e Umidade Relativa do ar 90%). O enraizamento foi avaliado após 30 dias. As estacas com folhas e sem folhas tiveram 100% e 57% de enraizamento, respectivamente, independente da concentração de IBA. O comprimento e o número de raízes nas estacas com folhas foram maiores que às estacas sem folhas em todas as concentrações de IBA. A brotação das estacas com folhas e sem folhas foi de 97,7% e 2,2%, respectivamente. A presença de folhas apicais aumenta o enraizamento de estacas de coleus e o uso de IBA é desnecessário para a propagação da espécie.

Palavras-chave: *Plectranthus scutellarioides*, auxina, ornamental, rizogênese, propagação vegetativa.

1. INTRODUCTION

Plectranthus scutellarioides (L.) R. Br., whose scientific synonyms are *Coleus blumei* Benth. (SYTAR et al., 2015) and *Solenostemon scutellarioides* (L.) Codd (YOKO et al., 2015), is popularly known as coleus (DATILES and ACEVEDO-RODRÍGUEZ, 2014).

This species belongs to the Lamiaceae family, characterized by the abundance of medicinal and ornamental plants. Coleus is mainly used for landscape composition (JUDD et al., 2009). In Ouro Preto, Minas Gerais State, Brazil, the plant is popularly known as Christ-blood, and is used for preparation of soothing teas by leaves infusion (MESSIAS et al., 2015). The species can also be consid-

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ered a hallucinogenic plant (DUKE, 2002). Another potential application for coleus plant is in phytoremediation of water contaminated by metals, since it showed remarkable capacity for aluminum extraction and accumulation (PANIZZA et al., 2011).

Coleus is characterized as perennial and vigorous foliage with upright, semi-upright, prostrate, semi-trailing, and trailing habits of growth (NGUYEN et al., 2008). It has been widely used as flowerbeds in landscaping gardening projects and has gained importance in the floriculture industry with the introduction of a large number of vegetatively propagated varieties with novel foliage colors and shapes (SAHU and DEWANJEE, 2012).

These genotypes are the result of breeding programs based on the genetic instability of this species, which generates unstable colors and shapes through mutations that also obstruct standardized coleus seeds propagation (WEISS, 2002). Clonal propagation is an alternative to the commercial multiplication of coleus (HAMILTON et al., 2002). Among vegetative propagation methods, the use of stem cuttings has advantages such as homogeneity and the maintenance of stock plants characteristics (HARTMAN et al., 2011), such as leaf color pattern. This method is associated with the use of auxin-based plant growth regulator which stimulate adventitious rhizogenesis (OLIVEIRA et al., 2011). Indolebutyric acid (IBA) is a synthetic auxin used for rooting. It is recognized to promote homogeneity and speed in stem cuttings rooting, and, consequently, to reduce the time required to commercial plantlets production (HAN et al., 2009). However, the required plant growth regulator concentration vary, among other factors, according to the species, the stock plants genetic and physiological quality, and the type of cuttings (PIZZATTO et al., 2011).

An additional aspect that may influence stem cuttings rooting is the presence of leaves, since they are sources of auxins and rooting co-factors, that, being transported to cuttings bases, will act synergistically to promote adventitious rooting (FERREIRA et al., 2009). The present study aimed to evaluate the influence of apical leaves and the use of different IBA concentrations in coleus vegetative propagation through stem cuttings.

2. MATERIAL AND METHODS

Coleus plants (branches with leaves) were collected in a private residence garden in Curitiba, Parana state, Brazil (Coordinates 25°24'58.74 "S and 49°16'35.05" W) on August 7, 2017 (Brazilian winter). Stem cuttings with 6 cm long semi-hardwood were made with a bevel cut at the base and a straight cut at the apex, with or without keeping a pair of apical leaves. Subsequently, the stem cuttings were treated by immersion in 0.5% sodium hypochlorite solution for 10 minutes, and then washed in running water for 5 min.

After phytosanitary procedure, stem cuttings had their bases treated with indolebutyric acid (IBA) at the following concentrations: 0 (control), 1,000 or 2,000 mg L^{-1}. IBA application was done through immersion of cuttings bases in hydroalcoholic solutions (50% v v^{-1}) containing the above-mentioned concentrations during 10 seconds. In the control treatment, cuttings had their bases immersed in hydroalcoholic solution without IBA. After IBA treatments, stem cuttings were planted in plastic tubes filled with previously dampened fine granulometry vermiculite. Cuttings had half their length inserted into substrate. Tubes with cuttings were kept for 30 days in a greenhouse with intermittent misting (24 ± 2 °C and 90% air relative humidity).

After 30 days from planting, the following variables were measured: i) cuttings rooting percentage (living cuttings that emitted roots greater than or equal to 2 mm); ii) number of roots per cutting; iii) average length of the 3 largest roots per cutting; iv) living cuttings percentage (cuttings with living tissues but without rooting or calli formation); v) calli formation percentage (living cuttings without rooting and with undifferentiated mass of cells at the base); vi) cuttings sprouting percentage; and vii) cuttings leaf maintenance percentage (cuttings that kept the original apical pair of leaves).

The study was conducted under a completely randomized experimental design in a 3x2 factorial scheme (3 IBA concentrations x 2 types of cuttings), with 4 replications and 10 stem cuttings per plot, totaling 240 stem cuttings.

Variances were tested for their homogeneity by the Bartlett test and, when homogeneous, data were submitted to variance analysis (ANOVA). Subsequently, the variables with significant differences had their means compared by the Tukey test at 5% probability.

3. RESULTS AND DISCUSSION

The interaction between the types of cuttings and IBA concentrations was only significant for sprouting percentage ($p = 0.05$). The factors were independent for the other variables.

Rooting in coleus stem cuttings was not affected by IBA. However, the rooting percentages differed significantly according to the type of cuttings. Rooting of coleus cuttings with and without leaves was 100% and 57.3%, respectively (Table 1). These findings corroborate with the study published by Hamilton et al. (2002) on the effects of leaf area on coleus stem cuttings rooting. These authors concluded that leaf area kept on stem cuttings have an expressive effect on rooting quality.

Table 1. Results of variance analysis (ANOVA), rooting percentage, average number of roots per cutting, average length of three largest roots per cutting, living cuttings percentage, and sprouting percentages in coleus (*Plectranthus scutellarioides*) stem cuttings with presence (PL) or without (WL) apical leaves treated with different indolebutyric acid (IBA) concentrations.

Rooting (%)								
	Concentration of IBA mg L^{-1} (F-calculated: 1.24^{ns})							
Cutting type (F-calculated: 55.41**)	0		1,000		2,000		Mean	
PL	100.0		100.0		100.0		100.0	A
WL	45.0		60.0		67.0		57.3	B
Mean	72.5		80.0		83.5			
F-calculated for interaction cutting type x concentration of IBA: 1.24ns; C.V.: 17.91%								
Number of roots per cutting								
	Concentration of IBA mg L^{-1} (F-calculated: 5.45*)							
Cutting type (F-calculated: 64.16**)	0		1,000		2,000		Mean	
PL	13.0		15.0		20.0		16.0	A
WL	4.0		6.0		7.0		5.7	B
Mean	8.5	a	10.5	a	13.5	a		
F-calculated for interaction cutting type x concentration of IBA: 0.78ns; C.V.: 28.78%								
Average length of three largest roots (cm)								
	Concentration of IBA mg L^{-1} (F-calculated: 1.54^{ns})							
Cutting type (F-calculated: 59.72*)	0		1,000		2,000		Mean	
PL	3.0		2.5		2.4		2.6	A
WL	0.6		1.6		0.9		1.0	B
Mean	1.8		2.05		1.65			
F-calculated for interaction cutting type x concentration of IBA: 5.10ns; C.V.: 27.73%								
Living cuttings (%)								
	Concentration of IBA mg L^{-1} (F-calculated: 1.24^{ns})							
Cutting type (F-calculated: 55.41**)	0		1,000		2,000		Mean	
PL	0.0		0.0		0.0		0.0	B
WL	55.0		40.0		33.0		42.7	A
Mean	27.5		20.0		16.5			
F-calculated for interaction cutting type x concentration of IBA: 1.24ns; C.V.: 65.82%								
Sprouting (%)								
	Concentration of IBA mg L^{-1} (F-calculated: 0.66^{ns})							
Cutting type (F-calculated: 4056.86**)	0		1,000		2,000		Mean	
PL	100.0	Aa	98.0	Aa	95.0	Aa	97.7	
WL	0.0	Bb	0.0	Bb	6.7	Ba	2.2	
Mean	50.0		49.0		50.9			
F-calculated for interaction cutting type x concentration of IBA: 5.62*; C.V.: 7.35%								

* means followed by the same lowercase letter in the rows and capital letter on the columns do not different by Tukey’s test at 5% probability.

The importance of leaves in stem cuttings rooting is related to the production of carbohydrates by photosynthesis, which will provide a source of energy, necessary for roots emission (VEIERSHOV, 1998).

Considering the average number of roots per cuttings, the presence of leaves also promoted better results. On average, 16 roots per cutting were verified in cuttings with apical leaves, a number almost three times higher than the 5.7 roots in leafless ones (Table 1). This behavior can be attributed to the fact that apical leaves are a major source of endogen auxin, a hormone directly related to adventitious rooting in stem cuttings (HARTMANN et al., 2011). Number of roots was affected by IBA concentrations (Table 1). Better results were observed at 2,000 mg L^{-1} IBA (Table 1). IBA is a plant growth regulator widely used to stimulate stem cuttings rhizogenesis when endogen auxin production is insufficient for rooting and roots development (FRAGOSO et al., 2017). In coleus micropropagation, similarly, nodal fragments treated with IBA presented early rooting and an increase in the number of roots per propagule (SAHU and DEWANJEE, 2012).

Stem cuttings with apical leaves also presented longer roots compared to leafless, both not influenced by IBA concentrations (Table 1). This behavior confirms that rooting and root system quality in coleus stem cuttings are highly associated with the presence of leaves, probably because the leaves provide reserves, hormones and co-factors that are not in sufficient concentration solely in coleus stems.

No mortality was observed in any treatment. Similarly, regardless IBA concentrations or presence/absence of leaves, no calli formation occurred on cuttings. This behavior evidences that coleus stem cuttings undergo direct rhizogenesis, i.e., rooting in this species does not require previous formation of undifferentiated mass of cells in cuttings base.

Considering the living cuttings (living tissues with no roots or calli), this was a behavior only observed in leafless cuttings, since all cuttings with leaves have rooted. On average, 42.7% of leafless cuttings remained alive with no roots nor calli formation.

For the leaf maintenance, on average, 45.8% of the cuttings kept the original pair of leaves until the evaluation, regardless IBA concentrations (data not shown).

Regarding the sprouting percentage, cuttings with leaves had the best results, and did not present variations according to IBA concentrations. On average, 97.7% of the cuttings with leaves have sprouted. On leafless cuttings, only a small percentage of sprouting was observed when 2,000 mg L^{-1} IBA was applied. No sprouting was observed in leafless cuttings submitted to the control treatment or treated with 1000 mg L^{-1} IBA (Table 1).

The sprouting in cuttings with leaves may also have been a factor influencing better rooting, since, according to Nicoloso et al. (1999), the new leaves produced during the rooting period are an essential feature for roots emission. The importance of leaves and new shoots in cuttings relies on the fact that these organs produce plant hormones, such as indoleacetic acid, and rooting cofactors, both essential for adventitious root formation in vegetative propagules (FACHINELLO et al., 2005).

The potential for coleus vegetative propagation is highly associated with the presence of leaves in stem cuttings, since it promoted better results for rooting, roots development and sprouting. IBA had little effect over rooting and sprouting in stem cuttings of this species. Probably the endogenous level of auxins in leaves provide enough stimuli for rooting in *P. scutellarioides* and, when transported to the cuttings base, promote rooting without the requirement for exogenous auxin application. The lack of IBA effect on leafless cuttings, though, may also suggest that not only auxin is a limiting factor for rooting in coleus cuttings, but also rooting co-factors only produced by the leaves. The nature and quantitative importance of these co-factors, however, are still to be understood for this species.

4. CONCLUSIONS

The use of indolebutyric acid is not necessary to promote coleus stem cuttings rooting and sprouting. The presence of apical leaves on the cuttings is a fundamental feature to promote rooting, roots growth and sprouting in this species.

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AUTHORS CONTRIBUTIONS

A.C.B.: study conception and design, acquisition, analysis and interpretation of data, manuscript draft and review. **L.A.N.R.:** Study conception and design, acquisition, analysis and interpretation of data, manuscript draft and review. **E.N.G.:** Interpretation of data, manuscript draft and critical review. **K.C.Z.R.:** study conception and design, manuscript critical review.

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