

Production of *Acalypha wilkesiana* seedlings using stem cuttings

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Abstract

The objective of this work was to evaluate the rooting of cuttings of Acalifa (*Acalypha Wilkesiana*) with different concentrations of indolbutyric acid (IBA). The matrix plant from which the cuttings were taken, is planted as a hedge and located at the biology institute of the Federal Rural University of Rio de Janeiro in the city of Seropédica-RJ. The experimental design was completely randomized, in a 3x3 factorial, with three different concentrations of IBA (0, 1000 and 2000 mg L⁻¹ or 0, 2000 and 4000 mg L⁻¹ for 15 seconds) and three types of cuttings (herbaceous, semi-woody and woody), with three replications and ten cuttings per plot. The experiments were carried out at two different seasons, namely: autumn and winter /spring. The following were evaluated: percentage of live cuttings, rooted, with callus and sprouting, number of roots per cut, length of the largest root. *Acalypha wilkesiana* can be propagated by stem cuttings, whether woody, semi-woody or herbaceous cuttings. However, herbaceous cuttings have a lower percentage of survival and rooting. Also, it is a species of easy rooting through cutting of stems, dispensing the use of AIB. Stems collected in the autumn period yield greater efficiency and productivity in the propagation of Acalifa by cuttings.

Keywords: Acalifa, auxin, propagation, seedling production

Introduction

The propagation and use of ornamental plants has been following the growing interest of the population in improving the quality of life, increasing investment in landscaping environments and generating a demand for particular production techniques of different species. (Alencar & Cardoso, 2015; Loss et al., 2015).

Acalypha Wilkesiana (Euphorbiaceae), also known as Acalifa, turkey's crest, monkey's tail, has leaves of ornamental importance for being of various colors and shapes, being widely used in landscaping. It is a semi-woody, perennial shrub, with a height ranging from 1.5 to 3 m (Souza et al., 2006; Lorenzi, 2015). Acalifa is used in public gardens (Baratta Junior, 2007) and it is often used in ornamentation as a living fence or even alone.

In addition to this function, Oso et al. (2016) mention that the red leaves of Acalifa can be implanted in the management of parasitic nematodes in agricultural

crops. Nagamoto et al. (2011) also highlight the possibility of using Acalifa in granulated baits to control leaf-cutting ants. The propagation of this species can be carried out by stem cuttings (Lorenzi, 2015).

Growth regulators have been frequently used to improve the rooting of some species (Vernier & Cardoso, 2013; Santos et al., 2014; Spassin & Garcia, 2016; Ataíde et al., 2017). This strategy aims to accelerate root initiation, optimize the emission and vigor of the roots, standardize rooting and, therefore, increase the percentage of rooted cuttings, reducing the time for seedling production (Fachinello et al., 1995; Wendling & Dutra, 2017). Such benefits can provide even greater chances of survival and better support of the root system in the transplant phase to the definitive location (Loss et al., 2015; Souza et al., 2018).

Indolebutyric acid (IBA) is a synthetic auxin, considered one of the best rooting stimulators (Rios et al.,

2012; Loss et al., 2015; Wendling & Brondani, 2015; Souza et al., 2018). However, there are great difficulties in dealing with specific recommendations, since the adequate concentration varies, among other aspects, according to the species or cultivar, type of cutting used, the state of propagules maturation, the season of its collection and the form of IBA application (Singh et al., 2011; Peña et al., 2012; Pereira et al., 2012; Costa et al., 2015; Souza et al., 2020).

No works are found in the literature that report the success or failure of rooting cuttings from *Acalypha*. Thus, the objective of this work was to evaluate the rooting and sprouting of three types of cuttings (herbaceous, semi-woody and woody) of *Acalypha Wilkesiana* treated with different concentrations of IBA.

Material and methods

The experiments were carried out in a greenhouse in the Experimental Area of the Department of Plant Science of the Federal Rural University of Rio de Janeiro (UFRRJ), located at latitude 22° 45' S, longitude 43° 41' W Grw and altitude of 33 m, with the climate being included as type Aw in the Köppen classification.

Such experiments were conducted at two seasons of the year (autumn and winter/spring). The first experiment was conducted from April 13 to May 31, 2017 (autumn) and the second from September 3 to October 29, 2018 (winter/spring), using herbaceous, semi-woody and woody cuttings, standardized according to the degree of lignification and stiffness of the branches.

The plants used as matrices for both experiments compose a hedge located in the Department of Phytopathology of UFRRJ in the city of Seropédica-RJ. Healthy branches, about 0.80 m long, were removed from these matrices. After this step, the branches were transported to the experiment site and prepared for cutting. Thus, these were cut in a bevel to obtain three cuttings measuring 0.15 m and standardized to have at least three buds per cuttings, the cut being made according to their position in the branch: subapical cuttings that are tenderer (herbaceous), basal cuttings (woody) and medium cuttings with intermediate characteristics to the others (semi-woody). For all cuttings, the leaves were carefully removed. The cuttings were disinfected by rapid immersion in sodium hypochlorite (1%) for 1 minute, followed by triple washing in distilled water. Subsequently, their lower extremities were treated with IBA solution at concentrations of 0, 1000 and 2000 mg L⁻¹ (autumn) and 0, 2000 and 4000 mg L⁻¹ (winter/spring) for 15 seconds for both seasons. The solutions were obtained by diluting the IBA in 50% alcohol to obtain the

desired IBA concentrations. For the control (0 mg AIB L⁻¹ concentration), distilled water was used during the same immersion time. The total number of *Acalypha* cuttings used in each experiment was 270, with 90 cuttings for each developmental stage mentioned above. The cuttings were taken to the beds located in the shade and filled with coarse washed sand, their bases being fixed in this inert substrate at a depth equivalent to about 1/3 of their size and irrigated by misting (10 seconds every 8 minutes), remaining under these conditions for a period of 47 and 56 days for the first and second seasons of the experiments, respectively.

The experimental design was randomized blocks, in a 3x3 factorial, with three different concentrations of IBA (0, 1000 and 2000 mg L⁻¹ in autumn) and (0, 2000 and 4000 mg L⁻¹ in winter/spring) and three types of cuttings (herbaceous, semi-woody and woody) in the two seasons, with three replications and ten cuttings per plot. Thus, each experiment had 27 plots, consisting of 10 cuttings each, totaling 270 cuttings. The stem cuttings were arranged in blocks with nine rows each, containing 10 cuttings per row, with random treatment (spacing was ± 3 cm between cuttings and rows). Each treatment (0, 1000 and 2000 ppm or 0, 2000 and 4000 ppm of IBA) consisted of 30 cuttings (3 lines with 10 cuttings), in which each line was a repetition.

After 47 and 56 days for the first and second experiments, respectively, the percentages of live cuttings, rooted cuttings, cuttings with shoots, cuttings with calluses, number of roots per cutting and length of the largest root were evaluated.

For all data, the evaluation of data normality (Lilliefors) was performed. Subsequently, the results were submitted to analysis of variance with the application of the F test and compared by the Tukey test ($p < 0.05$), using the SISVAR 5.3 program, from the Federal University of Lavras.

Results and discussions

It was found that in none of the treatments and at different season, cuttings with calluses were identified, indicating that the initiation of root primordia is not related to calluses. When analyzed separately, the type of cuttings factor (TE) showed a statistical difference for the variables percentage of live cuttings (EV), cuttings with shoots (EB) and number of roots (NR) (Table 1). In this sense, woody and semi-woody cuttings presented higher EV and EB in relation to herbaceous type cuttings (Table 1). This may have occurred due to the accumulation of reserves in cuttings with larger diameters (woody and semi-woody), favoring and costing the rapid emission of

shoots.

Fachinello et al. (1995) mentioned that cuttings with larger diameters have higher levels of reserve substances, favoring rooting, but may also have high rates of shoots, impairing root formation. This may have occurred in this work, as the herbaceous type cuttings presented a higher NR compared to semi-woody and woody cuttings (Table 1). This is probably related to the fact that the highest concentrations of free auxin in plants are found in the apical meristems of the shoot and young leaves, as they are the main sites of biosynthesis of this

hormone (Taiz & Zeiger, 2006).

The AIB concentration factor (CA) in the autumn season resulted in a statistical difference only for the variable root number (NR), verifying that the concentration of 2000 mg L⁻¹ of AIB provided a higher NR in relation to the control (0 mg AIB L⁻¹) (Table 1). This result raised the hypothesis that higher concentrations of IBA could have a significant effect. Thus, for the winter/spring season experiment, a higher dose was tested, as can be seen in this article.

Table 1. Percentage of live (EV), rooted (EE) and sprouting (EB) cuttings, number of roots (NR) and longest root length (CR) of three types of *Acalypha Wilkesiana* cuttings, treated with different concentrations of IBA, at 47 days after experiment deployment in autumn.

Treatments	EV	EE	EB	NR	CR
	----- % -----			-- cm --	
Herbaceous	85.55 #b	82.22 a	83.33 b	27.17 a	5.58 a
Semi-woody	95.55 a	92.78 a	95.55 a	19.79 b	7.31 a
Woody	95.55 a	87.78 a	95.55 a	18.15 b	6.08 a
0 mg L ⁻¹ of AIB	90.00 a	86.67 a	90.00 a	19.74 b	5.91 a
1000 mg L ⁻¹ of AIB	90.00 a	82.22 a	88.89 a	21.00 ab	5.99 a
2000 mg L ⁻¹ of AIB	96.67 a	93.89 a	95.55 a	24.37 a	7.07 a
Types of cuttings (TE)	3.86 *	2.21 ns	7.93 **	15.35**	3.28 ns
AIB concentrations (CA)	1.71 ns	2.71 ns	2.03 ns	3.81 *	1.72 ns
TE x CA	0.43 ns	2.19 ns	0.36 ns	17.56 **	7.58 **
CV (%)	9.56	12.16	8.22	16.94	23.42

Means followed by the same letters in the column do not differ statistically by Tukey's test, at 5% probability. ns - Not significant. * and ** significant by the F test, at 5 and 1% probability, respectively.

It was found that for the autumn season there was a significant interaction between the factors "types of cuttings" and "IBA concentrations" for the variables root number (NR) and longest root length (CR) (Table 1 and 2). The highest concentration of IBA tested (2000 mg L⁻¹ IBA) increases root number in herbaceous cuttings and root length in semi-woody cuttings (Table 2). However, this high concentration of IBA (2000 mg L⁻¹ of IBA) causes a shorter root length when compared to the control (0 mg L⁻¹ of IBA) (Table 2). These results show that the application of the highest concentration of IBA in the woody cutting can become phytotoxic, due to this type of cutting has a larger diameter and, consequently, has a greater accumulation of the phytohormone. On the other hand, herbaceous and semi-woody cuttings, due to their lower accumulation of reserves than woody cuttings, responds more efficiently to the application of IBA for these variables, indicating the absence of this same phytotoxic effect (Table 2). This can even be better seen when evaluating the types of cuttings within each dose, verifying lower root number (concentration of 1000 and 2000 mg L⁻¹ of IBA) and lower length of the largest root (2000 mg L⁻¹ of IBA) of the woody cuttings in relation to the herbaceous (Table 3). This is even more evident within the control (0 mg L⁻¹ of IBA), in which the woody type cuttings had a longer root than the semi-woody and

herbaceous cuttings (Table 2).

Sarzi & Pivetta (2005), evaluating the effect of seasons and different concentrations of IBA (0, 500, 1000, 1500, 2000 ppm) on the rooting of subapical cuttings of *Rosa spp.* (Rosaceae), found rooting only in spring and summer, demonstrating a correlation with the time of year of staking and suggesting that this is a determinant variable for productive success. Similarly, Zem et al. (2015), working with the species *Drimys brasiliensis*, also demonstrated that the season of collection and the type of cutting interfere in the percentage of survival and rooting. In view of the above, it can be speculated that this relationship could also occur for *Acalypha*, because as can be seen when comparing the data in Table 1 x Table 3, the results of the main parameters indicate a reduction in percentages for cuttings collected and rooted during test in winter/spring, the only exception being those of the woody type.

The season of the year can influence root induction due to nutrient reserves in cambium tissues and cambium activity, as well as the distribution of endogenous auxins in cuttings (Ohland et al., 2009; Zem et al., 2015). Thus, the effect of exogenous auxins can vary at different times of the year, presenting an optimal concentration that varies between species, promoting or inhibiting the rooting process (Fischer et al., 2008), and

may become phytotoxic in some cases (Zuffellato -ribas & Rodrigues, 2001). In addition, the presence of phenolic compounds, possibly from the group of monophenols, can cause auxin degradation and negatively interfere with rooting induction, as well as the insufficiency of some auxin cofactors at a certain season of year (Lima et al., 2011).

There was no significant interaction between the factors "cuttings types" and "AIB concentrations" for any

of the variables analyzed (Table 3). When these factors are evaluated separately, it can be seen that was a statistical difference for the type of cuttings factor, where higher EV, EE, EB and NR were obtained in woody type cuttings in relation to herbaceous ones (Tables 3). It is also noteworthy that, for these variables, semi-woody cuttings obtained better results than herbaceous cuttings, as was expected due to greater lignification and reserves (Tables 3).

Table 2. Statistical breakdown of the effect of the interaction between type of cuttings and IBA doses in terms of root number and length of the largest root.

Types of cuttings	AIB concentrations		
	0	1000 mg L ⁻¹	2000
	Number of roots		
Herbaceous	18.17 aB	22.85 abB	40.40 aA
Semi-woody	17.83 aA	24.60 aA	16.95 bA
Woody	23.23 aA	15.57 bA	15.67 bA
	Longest root length (cm)		
Herbaceous	5.02 bA	4.70 aA	7.01 abA
Semi-woody	4.76 bB	7.20 aAB	9.98 aA
Woody	8.19 aA	5.82 aAB	4.23 bB

Means followed by equal letters, lowercase in the columns and uppercase in the row do not differ statistically by Tukey's test, at 5% probability.

Table 3. Percentage of live (EV), rooted (EE) and with shoots (EB) cuttings, number of roots (NR) and longest root length (CR) of three types of *Acalypha Wilkesiana* cuttings, treated with different concentrations of IBA, at 56 days after implantation of the experiment in winter/spring.

Treatments	EV	EE	EB	NR	CR
	----- % -----			-- cm --	
Herbaceous	48.88 #c	48.88 c	48.88 c	20.68 c	9.38 a
Semi-woody	72.22 b	72.22 b	72.22 b	28.15 b	9.05 b
Woody	98.88 a	98.88 a	98.88 a	29.62 a	9.03 b
0 mg L ⁻¹ of AIB	80.00 a	80.00 a	80.00 a	26.32 a	9.20 a
2000 mg L ⁻¹ of AIB	78.88 a	78.88 a	78.88 a	28.15 a	9.14 a
4000 mg L ⁻¹ of AIB	61.11 a	61.11 a	61.11 a	29.62 a	9.12 a
Types of cuttings (TE)	20.59 **	20.59**	20.59 **	1263.10**	28.66**
AIB concentrations (CA)	3.69 ns	3.69 ns	3.69 ns	1.35 ns	1.08 ns
TE x CA	0.65 ns	0.65 ns	0.65 ns	0.67 ns	0.66 ns
CV (%)	22.56	22.56	22.56	1.54	1.20

Means followed by the same letters in the column do not differ statistically by Tukey's test, at 5% probability. ns - Not significant. * and ** significant by the F test, at 5 and 1% probability, respectively.

Regardless of the two tested times, it appears that higher percentages are related to woody cuttings, which reinforces the hypothesis of greater diameter and accumulation of reserves, boosting survival and rooting. According to Oliveira et al. (2001), in some species, the size and diameter of the cuttings can influence the rooting even of easy-to-root species. Souza et al. (2006), when evaluating the influence of substrates on the production of seedlings of *Acalypha* in Ilha Solteira-SP, used woody cuttings and verified that the species presented 1 to 2 shoots at 30 days and 12 to 14 roots at 60 days, concluding that regardless of the substrate used, *Acalypha* seedlings are suitable for the market between 30 and 60 days.

On the other hand, the lack of response to the

application of different concentrations of IBA reinforces that *Acalypha wilkesiana* must contain sufficient endogenous content of auxin and rooting co-factors, in addition to the absence or low concentration of phenolic compounds and other inhibitory substances, which is why of the expressive percentage of rooting, even when only treated with water. The ability of a cutting to form roots varies with the species or cultivar and results from the interaction of several factors such as the presence of rooting cofactors, phenolic compounds, the presence of anatomical barriers, and not just the genetic potential (Lima et al., 2011). In view of this, the species can be classified as "easy rooting" that its cutting tissues provide all the substances (mainly auxins) necessary for the formation of root primordia, dispensing with exogenous treatments.

Conclusions

Acalypha wilkesiana can be propagated by stem cuttings, whether woody, semi-woody or herbaceous cuttings. However, herbaceous cuttings have a lower percentage of survival and rooting.

The species is easy to root through stem cuttings, dispensing the use of IBA.

Stems collected in the winter/spring period yield greater efficiency and productivity in the propagation of seedlings by cuttings.

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Conflict of Interest Statement: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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