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# Growth regulators in the rooting of sun poinsettia (*Euphorbia pulcherrima* Willd. ex Klotzsch, Allg. Gartenzeitung) Valenciana variety

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#### ABSTRACT

**Objective**: To improve the asexual propagation of the sun poinsettia (*Euphorbia pulcherrima*) Valenciana variety using growth regulators and monitoring the position of the buds on the stem.

**Methodology**: We compared root length, shoot length, and the number of roots, shoots, and leaves between control plants and plants with five different concentrations of three growth regulators (indole butyric acid, naphthaleneacetic acid, and gibberellic acid). We also evaluated the effect of the position of the axillary or vegetative buds on the stem on the same parameters. The data were analyzed by analysis of variance and multiple comparisons of means.

**Results**: The treatments with the highest root length, shoot length, and number of roots, nodes, and leaves were 2 A (AIB 200 mg L<sup>-1</sup>, basal part), 3 A (AIB 300 mg L<sup>-1</sup>, basal part), and 3 B (AIB mg L<sup>-1</sup>, intermediate part).

Implications: This work helps solve a technical issue of sun poinsettia production in Mexico.

**Conclusions**: The application of indolbutyric acid at concentrations of 200 mg  $L^{-1}$  and 300 mg  $L^{-1}$  in stem cuttings of the intermediate and base promotes increased root and aerial development.

Keywords: auxins, ornamental plant, growth regulators.

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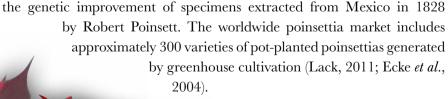
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#### INTRODUCCIÓN

The poinsettia (*Euphorbia pulcherrima*), also known as nochebuena, cuetlaxóchitl, Christmas flower, cardinal star, or federal star; is a plant native of Mexico, naturally distributed from Sinaloa to Guatemala. This species is cultivated worldwide thanks to



In Mexico, the poinsettia is commonly cultivated outdoors, growing bushily in orchards or family gardens and decorating with its colorful inflorescence. This species is a shrub with a height of up to 5 m and an inflorescence in the form of a cyathium, which consists of a female flower without petals



or sepals surrounded by individual male flowers (Ecke *et al.*, 2004). Nowadays, nine sun varieties are registered in Mexico's National Catalogue of Plant Varieties (CNVV) of the National Service for Seed Inspection and Certification (SNICS). Most of these varieties were obtained by horticulturists from the State of Morelos, Mexico, through the selection, breeding, and modification of creole poinsettia (García *et al.*, 2011; Colinas *et al.*, 2014) (Figure 1).

In particular, the Valenciana variety is distinguished by abundant bracts of different sizes, which form a kind of crest on the deformed red cyathia; this variety is the most commonly cultivated in Morelos. Horticulturists propagate it by cuttings, which are directly introduced into a substrate rich in organic matter without applying any hormonal treatment. This propagation procedure causes insufficient rooting, which, together with the scarce technical information regarding poinsettia cultivation, represents a problem for horticulturists.



Figure 1. Sun varieties registered in Mexico (A=Orejona, B= Juan Pablo, C=Valsu, D=Belén, E=Valenciana, F=Corona, G=Rehilete, H=Tete, I=Amanecer Navideño (Adapted from Colinas *et al.*, 2014).

The present work aims to offer an alternative for the cultivation of the Valenciana variety of poinsettia in Morelos. We used growth regulators and monitored the position of the buds on the stem to obtain more roots, thus improving the asexual propagation of the plant.

#### MATERIALS AND METHODS

Stems of sun poinsettia (*Euphorbia pulcherrima*) Valenciana variety were collected in March 2020 from the Germplasm Bank of the Universidad Autonoma Chapingo, Mexico (Figure 2), which is located at 19° 29' 23" west and 98° 53' 37" north.

The stems were cut into segments containing 2 to 3 buds of approximately 20 cm in length, taking care that the cut was clean, the stem was unbroken, and at least three buds were maintained after the cut. Once the semi-hardwood cuttings were obtained, they were grouped into three groups depending on the position of the cut in the stem (A=base, B=middle part, and C=upper or apical part of the stem), (Figure 3).

Aqueous solutions containing powdered reagent-grade growth regulators were prepared, dissolved with alcohol, and volumetrically diluted in water; the pH was adjusted to 5.7. The semi-hardwood cuttings were placed for 24 h in the solutions with different concentrations of the growth regulators. The cuttings maintained their polarity with respect to the base, and half of the cutting was placed in the solution. Once the time had elapsed, 12 cuttings from each treatment were placed in plastic boxes with a substrate containing peat moss+agrolite in a 2:1 v/v ratio. Half of the cutting was kept in the substrate to avoid dehydration during the rooting period.

The semi-hardwood cuttings were maintained in a glass greenhouse with temperatures ranging between 38 and 9 °C. The cuttings were irrigated two to three times per week



Figure 2. Poinsettia material in the germplasm bank of the UACh.

	A		В			C		bud 	
o_	0	0	0	0	0	0	0	04	
base			middle			upper			

Figure 3. Semi-hardwood cutting according to stem position.

until evaluation three months later. Five treatments containing different concentrations of indolbutyric acid (IBA), naphthalenic acid (NAA), and gibberellic acid (GA<sub>3</sub>) were evaluated along with a control. The treatments were as follows: 1) AIB 100 mg L<sup>-1</sup>, 2) AIB 200 mg L<sup>-1</sup>, 3) AIB 300 mg L<sup>-1</sup>, 4) AIB 100 mg L<sup>-1</sup>+ANA 100 mg L<sup>-1</sup>, 5) AIB 100 mg L<sup>-1</sup>+ANA 100 mg L<sup>-1</sup>+AG<sub>3</sub> 100 mg L<sup>-1</sup>, and 6) no growth regulator (control). The parameters evaluated were root number, root length, node number, shoot length, and leaf number. The results were analyzed by analysis of variance; multiple comparisons of means were performed using Tukey's honest significant difference with significance level at 5%, and the general linear model theory was used with the GLM procedure of SAS (SAS Institute, 2015).

#### RESULTS AND DISCUSSION

After three months, the treatments 2 A (AIB 200 mg L<sup>-1</sup>, basal part of the semi-hardwood cuttings) and 3 A (AIB 300 mg L<sup>-1</sup>, basal part) generated the greatest amount of roots; while 5 A and 5 B (AIB 100 mg L<sup>-1</sup>+ANA 100 mg L<sup>-1</sup>+AG<sub>3</sub> 100 mg L<sup>-1</sup> in the basal and middle part, respectively) generated the least amount of roots (Table 1). These results suggest that the combination of AIB with other growth factors was not efficient for root generation; however, the application of AIB alone yielded a higher root production (Figure 4).

The application of auxin-based rooting agents promotes root production in poinsettia semi-hardwood cuttings. Since this species has a hollow stem structure, it is difficult to apply powdered rooting agents to the small contact surface; so the application in liquid form guarantees a better response. Similar results were found by Amri et al. (2010), who showed that the use of AIB promoted the rooting of *Dalbergia melanoxylon* (granadilla). Other ornamental species where AIB has been used successfully are *Hibiscus rosa-sinensis* L., *Conocarpus erectus*, *Rosa caninna* L., *Carisa carandas* L., *Streptosolen jamesonii*, and *Solidago* 



**Figure 4.** Comparison of the worst treatment (5 B,  $100 \text{ mg L}^{-1} + \text{ANA } 100 \text{ mg L}^{-1} + \text{AG}_3 100 \text{ mg L}^{-1}$ , middle part) against the best one (3 A, AIB  $200 \text{ mg L}^{-1}$ , basal part) for rooting of *Euphorbia pulcherrima* Valenciana variety.

canadensis L. (Abdel-Rahman et al., 2020; Tanuja and Rana, 2018; Pêgo et al., 2019). AIB, a synthetic auxin, is commonly applied to various woody species for root generation because it regulates cell division, expansion, and differentiation (Frick and Strader, 2018). Commercial AIB is found alone or combined with other chemicals (Ludwig-Müller, 2000), making it easy to implement in horticulture; some examples of commercial AIB that could be used by horticulturists in Morelos include Radix<sup>®</sup>, Raizone<sup>®</sup>, Rootex<sup>®</sup>, among others.

Regarding the position of the buds, Table 1 shows that those located in the basal and middle part of the cutting generated a greater number of roots than those in the upper or apical part. These results are mainly because the basal and middle parts of the stems contain higher carbohydrate storage, a higher capacity for adventitious root formation, more organogenic activity, and higher micronutrient content (Zalesny *et al.*, 2003; Tsafouros *et al.*, 2019). Basal and mid-rooting has been reported in Rootpac-R and Myrobalan 29C compatible rootstocks in peach, nectarine, almond (Tsafouros *et al.*, 2019), sandalwood (Tate and Page, 2018), and granadilla (Amri *et al.*, 2010).

The treatments with the greatest root length were 2 A (AIB 200 mg L<sup>-1</sup>, basal part) and 3 A (AIB 300 mg L<sup>-1</sup>, basal part); while the lowest lengths were found in 5 A, 5 B, 5 C (AIB 100 mg L<sup>-1</sup>+ANA 100 mg L<sup>-1</sup>+AG<sub>3</sub> 100 mg L<sup>-1</sup> basal, middle and upper part, respectively) and 6 C (control, upper part) (Table 1).

**Table 1**. The effect of growth regulators on rooting and shoot development in semi-hardwood cuttings of sun poinsettia (*Euphorbia pulcherrima*) Valenciana variety.

Treatment		Number of roots	Root length (cm)	Number of buds	Number of leaves	Shoot length (cm)
1	A	18.08 abcd	12.50 abc	11.17 ab	14.00 a	57.75 a
	В	12.17 cd	6.17 abcd	9.25 ab	9.08 abcd	27.56 bcdefg
	С	5.50 d	5.71 abcd	8.67 ab	6.58 cde	14.54 fg
2	A	28.25 abc	13.58 a	13.75 a	11.33 abcd	46.25 abcd
	В	19.00 abcd	8.83 abcd	9.50 ab	9.83 abcd	31.25 abcdef
	C	5.83 d	4.42 dc	5.25 bcd	6.83 bcde	18.00 efg
3	A	34.33 a	13.13 ab	13.08 a	10.33 abcd	54.17 ab
	В	28.08 abc	12.88 abc	13.42 a	11.92 abc	48.83 abc
	С	7.25 d	8.08 abcd	8.33 abc	7.33 bcde	20.58 defg
4	A	32.25 ab	12.17 abc	5.42 bcd	7.08 bcde	24.75 cdefg
	В	14.25 bcd	6.46 abcd	2.58 cd	2.50 e	6.50 fg
	С	10.17 cd	4.79 bcd	2.17 d	1.83 e	4.00 g
	A	3.58 d	2.94 d	1.83 d	5.50 de	8.83 fg
5	В	1.58 d	1.96 d	1.58 d	2.75 e	3.92 g
	С	1.67 d	0.71 d	1.75 d	1.75 e	0.94 g
6	A	8.92 d	8.33 abcd	12.67 a	12.67 ab	44.17 abcdef
	В	6.92 d	7.90 abcd	11.83 a	12.25 abc	33.04 defg
	С	3.00 d	1.65 d	11.00 ab	8.92 abcd	20.58 defg
MSD		18.38	8.46	6.06	5.87	27.20

MSD=Minimum Significant Difference. Different letters in the same line denote significant differences (Tukey,  $\alpha$ =0.05), n=12.

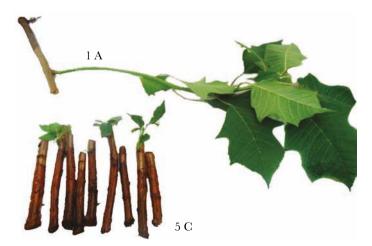
The results obtained in root length coincide with those obtained in root formation, again suggesting that the effectiveness of AIB is better without adding another growth regulator. AIB is known to promote root elongation and development in various crops (Frick and Strader, 2018); however, despite its use in agriculture since the 1950s, its effectiveness depends on the formulation, concentration, and species evaluated (Sisaro and Hagiwara, 2016). Ignoring the proper concentration of AIB can cause economic losses to producers (Ludwig Müller, 2000); therefore, the methodology proposed in the present experiment could increase the percentage of rooting and the length of roots generated in the cuttings.

The capacity of stem cell dedifferentiation and redifferentiation also determines root formation in different species (Amri *et al.*, 2010; Rout, 2006). In this sense, physiologically mature tissues have a lower rooting capacity than juvenile material (Aparicio *et al.*, 2014). In sun poinsettia variety Valenciana, the selection of stems of woody consistency and dark brown coloration generated few roots (pilot study, results not shown); therefore, we selected only those of semi-woody consistency and light brown color.

We also evaluated the development of the aerial part of the cuttings. The greatest shoot length was found in the 1 A treatment (AIB 100 mg  $L^{-1}$  basal part), while the 5 C treatment (AIB 100 mg  $L^{-1}$ +ANA 100 mg  $L^{-1}$ +AG<sub>3</sub> 100 mg  $L^{-1}$  upper or apical part) gave the shortest length (Figure 5).

Regarding the number of nodes, the best treatments were 2 A (AIB 200-basal part), 3 A (AIB 300 basal part), and 3 B (AIB intermediate part); in contrast, the treatments with the lowest number of nodes were 5 B (100 mg  $L^{-1}$ +ANA 100 mg  $L^{-1}$ +AG3 100 mg  $L^{-1}$  upper part) (Table 1).

The treatments with the highest number of leaves were 1 A (AIB 100 mg L<sup>-1</sup> basal part), 3 A (AIB 300 mg L<sup>-1</sup> basal part), and 3 B (AIB 300 mg L<sup>-1</sup> middle part). These results can be explained by the position of the cuttings in the stem: thicker cuttings should have a higher concentration of cytokinins and more carbohydrates due to their proximity to the basal part (Oinam *et al.*, 2011; Rana and Sood, 2011). The development of the aerial



**Figure 5**. Shoot length in semi-hardwood cuttings of *Euphorbia pulcherrima* var. Valenciana in 1 A (AIB 100 mg  $L^{-1}$  basal part) and 5 C (AIB 100 mg  $L^{-1}$ +ANA 100 mg  $L^{-1}$ +AG<sub>3</sub> 100 mg  $L^{-1}$  upper or apical part).

part, including the length of shoots and number of leaves, depends on two factors: first, the carbohydrate reserves contained in the stem, and, later, on the nutrients and water that are transported by the newly formed roots (Verwijst *et al.*, 2012). Thus, when the stems are cut, the upper part will stimulate the axillary buds, generating shoots. The shoots will first depend on the reserves of the cutting and, later, when the roots are formed, nutrients and water will feed them. Therefore, if the roots do not develop, the aerial part will die.

#### CONCLUSIONS

The treatments with the highest number of roots, nodes, and leaves, as well as greater root and shoot length, were 2 A (AIB 200 mg  $L^{-1}$ , basal part), 3 A (AIB 300 mg  $L^{-1}$ , basal part), and 3 B (AIB mg  $L^{-1}$ , intermediate part).

The growth regulator that gave the best development of roots and shoots was indol butyric acid (IBA) when applied to the base and intermediate part of the cuttings at the concentration of 200 mg  $L^{-1}$  and 300 mg  $L^{-1}$ .

The results obtained in the present experiment support that the application of IBA yields more effective rooting without the addition of other growth regulators. We recommend applying liquid IBA to the Valenciana variety poinsettia stems to improve current propagation methods.

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