DEVELOPING AIR LAYERING PRACTICES FOR PROPAGATION OF DRACAENA MARGINATA LAM. UTILIZING PHLOROGLUCINOL AND SEAWEED EXTRACT AS IBA-SYNERGISTS OR ALTERNATIVES

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> ABSTRACT: In an experiment laid out in completely randomized block design, eight experimental treatments representing all-possible combinations between indole-3-butyric acid potassium salt at 300 ppm (IBA), phloroglucinol at 300 ppm (PG) and seaweed extract (SWE) at 2 ml/l were evaluated for their effects on rooting of Dracaena marginata Lam. air layers. The research was conducted under shade house conditions of the Nursery of Ornamental Plants, Faculty of Agriculture, Assiut University, Egypt during 2018 and 2019 successive growing seasons. All treated air layers with IBA, PG and SWE each alone or in combinations recorded significant increases in rooting percentage comparing to untreated control air layers. The highest rooting percentages were found in air layers received 10 ml/air layer from PG alone or the IBA + SWE or IBA + PG + SWE without significant differences between them. Air-layers exposed to IBA alone or in combination with PG and/or SWE attained the most favorable root traits in terms of high number of roots with appropriate length and a balanced mass and volume. High carbohydrate concentrations positively correlated with rooting percentage and roots weight and volume. Studying correlations between rooting characteristics and the internal contents of root zone tissues of total carbohydrates and total phenolics exhibited that rooting percentage correlated positively with total carbohydrates percentage and negatively with total phenolics in root zone tissues. While, the relationships between the other root traits of succeeded layers and total carbohydrates and total phenolics contents in rooting zones did not attain any significant relations.

> Key words: Indoor plants, *Dracaena*, red-edge dracaena, auxin, vegetative propagation, seaweed extract, phloroglucinol.

INTRODUCTION

According to APG IV classification system (Angiosperm Phylogeny Group, 2009, the plant list, 2020), Dracaena marginata Lam., a synonym of Dracaena reflexa var. angustifolia Baker., belongs to family Asparagaceae (formerly family Agavaceae or Dracaenaceae). It is an evergreen shrub native to Madagascar. It is characterized by multiple stems topped by a rosette of narrow ribbon-like green leaves edged in purplish-red, from which it has gained its popular common name red-edge dracaena. Its tolerance to irregular watering and little maintenance requirements makes it a durable houseplant favored by both home growers and landscape architects (Gilman, 1999). In addition, dracaena plant is regarded



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efficient Developing means of propagating D. marginata is crucial for the commercial production of this species. Vegetative propagation by cutting and air layering with some difficulty has been reported for this species. From the available information, it is clear that the exogenous application of auxins, mostly IBA, has positive effect on rooting percentage and root characteristics (Attia et al., 2015). IBA is the most famous rooting auxin employed to stimulate rooting in a wide range of plant species (Preece, 2003). However, Baker and Wetzstein (1994) recommended applying low concentration of IBA in order to avoid the inhibitory effect of high IBA concentration on rooting. This is further supported by evidence that IBA is converted to IAA in such a way that it provides a continuous supply of IAA at proper concentrations for rooting (George, 2008).

Plant growth regulators with auxin-like effects as phloroglucinol (PG) and seaweed extract (SWE) can provide materials to stimulate rooting and plant development. Such substances have shown favorable effects on vegetative propagation, commonly *in vitro*, for many plant species (Nardi *et al.*, 2016). According to the available literature, there are no previous studies respecting the effect of PG and SWE on *D. marginata* propagation by air layering. Both of PG and SWE have shown auxin-like effects on plant growth and development, which vary with their concentration, application method and plant species (Craigie, 2011).

PG is a benzenetriol phenolic compound naturally occurs in plants, marine algae and in many microorganisms. PG and its derivatives have been applied, mostly together with auxin, to stimulate *in vitro* rooting of many plants (Teixeira da Silva *et al.*, 2013) and stem and rhizome cuttings (Giri and Tamta, 2013). The mechanism by which PG stimulate rhizogenesis is mainly attributed to its protective effects against auxin decarboxylation and against plant tissue oxidative stress (De Klerk *et al.*, 2011). This may explain the PG-auxin synergistic effect reported by several authors, although its effectiveness is strongly dependent on genotype (Teixeira da Silva *et al.*, 2013).

Likewise, SWE has simulative effects on plant growth and development. Such effects may be attributed to direct or indirect impacts of the detected phytohormones in SWE (Ghaderi Ardakani et al., 2019). Several authors (Gomes et al., 2018) reported SWE stimulant effects on rhizogenesis and rooting percentage of cuttings. In some cases, such as stem cuttings of *Physocarpus opulifolius*, the effects of SWE were comparable to IBA (Pacholczak et al., 2016).

Because the effect of seaweed extract and/or phlorglucinol on *D. marginata* rooting has not been previously studied, the current research endeavored to evaluate the potential efficiency of phloroglucinol and/or seaweed extract as IBA synergists or alternatives for rooting of *Dracaena marginata* air-layers.

MATERIAL AND METHODS

The present study was performed under conditions of shade house (60% shading) of the Nursery of Ornamental Plants, Faculty of Agriculture, Assiut University, Egypt during the two successive growing seasons of 2018 and 2019 aiming to evaluate effectiveness of phloroglucinol and/or seaweed extract as IBA synergists or alternatives on *Dracaena marginata* Lam. propagation by air layering.

Healthy, vigorous and uniform D. marginata Lam. plants well established in soil of greenhouse were used in this study. The selected plants were 180 to 200 cm in height and each plant had 3 to 4 branches, averaged 3 cm in diameter for each.

The experimental treatments were set as a simple experiment containing eight treatments in a randomized complete block design with three replicates for each treatment; each replicate contained 15 air

layers (15 plants). So, 45 plants were used for each treatment. The eight experimental treatments the possible were all combinations between three experimental materials. The three tested materials were indole-3-butyric acid potassium salt at 300 ppm concentration (IBA), phloroglucinol at 300 ppm (PG) and seaweed extract at a rate of 2 ml/l (SWE). IBA and PG materials were purchased from El-Gomhouria Co. For Drugs, Egypt, while SWE was received from Oligo-X®, Union for Agriculture Development (UAD), Egypt. Therefore, the eight tested treatments were IBA, PG, SWE, IBA + PG, IBA + SWE, PG + SWE and IBA + PG + SWE, beside the control treatment that received distilled water only without any material.

The procedures:

Distilled water was used for preparing the designed rates of the above mentioned materials, PG was dissolved in little ethanol alcohol prior to use the distilled water.

On March 1st for both experimental seasons of 2018 and 2019 air layering was performed. To conduct air layering, one branch per plant with average 3 cm diameter and containing the same number of leaves was selected; and distance of 120 cm from the newest unfurled leaf of the terminal end of the selected branch was measured, which was girdled by removing 2 cm ring of bark (air layering position). Mixture of peat moss + perlite at 1:1 (v/v) was used as rooting substrate. Layering was performed immediately after girdling by covering the girdled region with the wet rooting substrate. Then, wrapped with a transparent plastic subsequently bag. and covered with aluminum foil. The designed treatments were applied by injecting 10 ml of the corresponding material the to rooting substrate for each air-layer. The prepared airlayers were protected against any mechanical damage and from moisture losing. Two and half months after air layering performance, air-layered branches were detached from mother plants, and the detached rooted layers were replanted in 30 cm diameter plastic

pots filled with clay soil and were kept under plastic house conditions for additional six months.

Recorded Data:

1. Rooting and survival percentage:

detaching time of air-layered At branches from mother plants, on June 15th for the two tested seasons (2.5 months after air layering process), rooting %, roots number per air layer, root length (cm) and root volume (cm³) that determined using water displacement as well as fresh and dry weights of roots per air-layer (g) were recorded. While, on December 15th of the two experimental seasons (six months after replanting the detached rooted layers), survival percentage of the planted detached air layers was recorded.

2. Chemical determinations:

At detaching time of air-layered branches, on June 15th for the two seasons, samples of 3 cm from the basal portions of the root zones of the detached air layers were taken. They were oven-dried at 70 °C for 72 hours and then well ground to use for chemical determinations. Total percentage carbohydrates was calorimetrically determined using enthrone sulphuric acid method described by Fales (1951). Also, total phenolic contents as mg GAE/g dry weight was colorimetrically estimated using Folin-ciocalteau reagent (FCR) method according to Maliauskas et al. (2004).

Statistical analysis:

Obtained data were statistically analyzed using Statistix 8.1 analytical software and the means were compared using least significant difference (LSD) test at 5% level according to Gomez and Gomez (1984). Additionally, Pearson's correlation coefficients between the internal levels of total phenolics and total carbohydrates in roots zone on one hand and the determined rooting percentage and root traits of rooted air-layers on the other hand were calculated using Statistic 8.1 analytical software.

RESULTS AND DISCUSSION

Rooting percentage:

Data representing rooting percentage in Fig. (1) show that all IBA, PG and SWE each alone or in combinations significantly increased percentage of rooted air-layered shoots of D. marginata plant comparing to untreated control air-layers. During the two experimental seasons, rooting percentage ranged from 46.67 to 100% in treated layers compared to 11.11% that recorded in the control ones. superior The rooting percentages were found in air-layered shoots that received 10 ml of PG at 300 ppm, IBA at 300 ppm + SWE at 2 ml/l or IBA at 300 ppm +PG at 300 ppm +SWE at 2 ml/l per air layer. There were no significant differences between these three treatments. Such result was confirmed during the two experimental seasons. The stimulatory effect of IBA on rooting and root growth has been elucidated in several previous studies (Kasem and Abd El-Baset, 2014), revealing the effect of IBA in accumulating metabolites at the site of application leading to stimulation of cells division and elongation as well as roots formation. This is further supported by evidence that IBA is converted to IAA, in such a way it provides a continuous supply of IAA at adequate concentrations for rooting (George, 2008), a process that has been suggested to be critical for successful propagation (Frick and Strader, 2018). Similar effect has also been reported on air layers of Ficus elastic var. decora (Mousa, et al., 2019) and Ficus carica (Reddy et al., 2014). Additionally, increases in rooting percentages were occurred in tip-shoot cuttings of D. marginata treated with IBA at 1000 ppm (Attia et al., 2015) and in D. sanderiana cuttings treated with IBA at 2000 ppm (Pushpamali et al., 2004).

The PG stimulating effect on rooting has been reported under *in vitro* conditions. In plant tissue culture, it is frequently added together with auxin to the rooting media to stimulate rooting of many plant species such as apple cultivars (Dubranszki and da Silva, 2010) and *Jatropha curcas* (Daud *et al.*,

2013). Scarce information is available about PG effects on rooting in vivo. Giri and Tamta (2013) on rhizome cuttings of Hedychium spicatum noticed PG promoting effect on rooting. However, James (1979) ascribed the rooting stimulant effect of PG in the absence of auxin application in some species to the presence of adequate levels of endogenous auxins, which synergistically respond to the exogenous PG. IBA-PG synergism can be explained on the basis of the role of PG in protecting IAA, the active auxin converted from IBA. Another promotive effect of PG is alleviating oxidative stress, which facilitate improved regeneration (De Klerk et al., 2011). Several studies have indicated that some polyphenol compounds act as rooting co-factors during the root formation process, especially as protectors of IAA against oxidation. Hess (1969) reported that PG acts as auxin synergistic or auxin protector. A Moderate effect of SWE was observed on rooting percentage in the absence of IBA. This effect highly increased in the presence of IBA and/or PG. The stimulatory effect of SWE at intermediate concentration on rooting percentage was previously reported in Pinus patula cuttings comparing to the poor rooting in cuttings exposed to high levels (Jones and van Staden, 1997). Exposure of Passiflora actinia stem cuttings to brown SWE had positive effects on rooting (Gomes et al., 2018). Pacholczak et al. (2012) recorded similar observations on cuttings of two dogwood cultivars. In cases of stem cuttings of common ninebark (Physocarpus opulifolius), the effects of SWE on rooting of stem cuttings were comparable to IBA (Pacholczak et al., 2016). The mechanism of SWE stimulative effect on plant growth and development might be due to direct or indirect effects of its content of phytohormones (Ghaderi Ardakani et al., 2019). Many phytohormones have been detected in a wide range of seaweeds (Stirk and Van Staden, 2014), of which auxins are active components in vegetative the propagation of plants. This effect may indirectly occur due to other substances

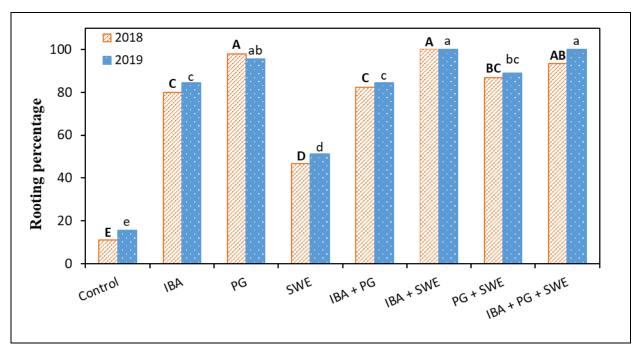


Fig. 1. Rooting percentage of *Dracaena marginata* air-layers as affected by different combinations of IBA, phloroglucinol (PG) and seaweed extract (SWE) during 2018 and 2019 seasons. Different capital and lower-case letters denote significant differences among the treatments in 2018 and 2019 seasons, respectively, using LSD test (p≤0.05).

contained in SWE that trigger plant signaling pathways.

Root characteristics:

Root characteristics of the succeeded air layers are recorded in Tables (1 and 2) and Fig. (2). From the presented data, it is generally evident that roots fresh weight per air layer (g) were significantly improved in response to the applied treatments comparing to untreated control air layers. Both fresh and dry weights of roots had similar trend, but values of dry weights did not exhibit significant differences. Air-layers that received PG, SWE, IBA + SWE or IBA + PG + SWE treatments resulted in the heaviest roots fresh weights/air layer during the two tested seasons comparing to control and the other treatments.

In terms of average root length (Table, 2 and Fig., 2), both of untreated air-layers and those treated with the combination of IBA+ PG + SWE produced the shortest roots. Using SWE alone or in combination with IBA induced generally the longest roots and, on average, 100% longer roots than either of the two treatments. Air-layers performance regarding number of roots responded significantly to the tested treatments (Table, 2 and Fig., 2). Layers treated with IBA alone or with PG + SWE showed the highest significant increases in roots number/layer during the two seasons comparing to the control and the other treatments. There were no significant differences between the two treatments. Average root volume in all treated air-layers, generally, was higher than those in untreated ones (Table, 2 and Fig., 2). SWE alone or its combination with both IBA and PG were the most effective treatments. followed by IBA alone then the combination of PG and SWE.

To comprehend the variations obtained in root characteristics, it is important to consider all of them together as each trait affects or is affected by one or more of the other traits. High roots volume noticed in airlayers treated with IBA alone or IBA + PG + SWE was accompanied with high values of root number and mass. However, high root

Cable 1. Roots fresh and dry weights/ air-layer (g) of Dracaena marginata Lam.	
rooted air-layers as affected by different combinations of IBA,	
phloroglucinol (PG) and seaweed extract (SWE) during 2018 and 2019	
seasons.	

Treatments				
	Fr	esh	D	ry
	2018	2019	2018	2019
Control	0.71	0.75	0.074	0.082
IBA	1.21	1.25	0.128	0.146
PG	1.61	1.66	0.144	0.151
SWE	1.56	1.60	0.159	0.173
IBA + PG	1.34	1.41	0.117	0.129
IBA + SWE	1.03	1.10	0.122	0.143
PG + SWE	1.00	1.00	0.102	0.116
IBA + PG + SWE	1.50	1.40	0.143	0.121
LSD (0.05)	0.49	0.22	NS*	NS

*NS denotes nonsignificant differences using ANOVA.

IBA and PG used concentrations were 300 ppm for each, while SWE rate was at 2 ml SW/ L. Each air layer received 10 ml of the corresponding material, and control treatment received 10 ml distilled water.

Table 2. Average root (length, volume and number per rooted air-layer) of Dracaenamarginata as affected by different combinations of IBA, phloroglucinol (PG)and seaweed extract (SWE) during 2018 and 2019 seasons.

Treatments	Root						
	Length (cm)		Number/air-layer		Volume (cm ³)		
	2018	2019	2018	2019	2018	2019	
Control	7.33	7.93	5.67	6.33	0.80	0.90	
IBA	11.00	11.50	9.33	9.67	1.67	1.83	
PG	12.00	12.50	6.00	5.67	1.37	1.63	
SWE	15.00	14.50	4.33	4.33	2.23	2.07	
IBA + PG	12.33	11.83	5.00	4.67	1.40	1.63	
IBA + SWE	15.00	15.50	3.67	4.33	1.20	1.33	
PG + SWE	13.00	13.50	9.00	9.33	1.63	1.53	
IBA + PG + SWE	7.33	9.17	4.67	5.00	2.17	2.00	
LSD (0.05)	1.46	1.44	1.17	1.15	0.56	0.35	

IBA and PG used concentrations were 300 ppm for each, while SWE rate was at 2 ml SW/L. Each air layer received 10 ml of the corresponding material, and control treatment received 10 ml distilled water.



Fig. 2. Rooted air-layers of *Dracaena marginata* Hort. as affected by different combinations of IBA, phloroglucinol (PG) and seaweed extract (SWE).

volume in air-layers treated with SWE was associated with root length and mass. It is generally accepted that this variation depends on other factors such as root thickness and density. However, several previous studies attained vigorous root system in cuttings or air-layers in response to the application of IBA (Mousa, *et al.*, 2019), PG (Daud *et al.*, 2013) and SWE (Gad and Ibrahim, 2018).

Survival percentage:

Despite the varying response of airlayers to the employed treatments in terms of rooting percentage and other determined root characteristics, yet survival percentage of the detached rooted air-layers (unpublished data) wasn't affected with the all-experimental treatments, and it was 100% in all treatments undependability of the survival percentage on the applied treatments.

Chemical determinations:

Data of total phenolics (mg GAE/g DW) and total carbohydrates (%) in tissues of air layers rooting zones in Table (3) revealed opposite directions for the two traits. Total phenolics values were significantly higher in tissues of untreated layers or those that received individual treatments of IBA or SWE than any of the other treatments. Generally, the values were lower in case of the combined treatments comparing to the individual ones, where the combination between the three substances, IBA + PG + SWE, inducing the lowest values of total phenolics. Association of high phenolics concentration in air-layers tissues with the

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Treatments		henolics E/g DW)	Total carbohydrates (%)	
	2018	2019	2018	2019
Control	5.07	5.70	16.49	15.53
IBA	4.74	4.88	20.51	18.06
PG	4.25	4.61	20.22	22.18
SWE	5.83	6.14	17.25	16.49
IBA + PG	4.14	4.05	18.98	18.06
IBA + SWE	3.59	4.40	20.01	19.01
PG + SWE	2.77	3.40	16.48	17.79
IBA + PG + SWE	2.61	3.57	20.81	22.48
LSD (0.05)	1.55	1.15	1.89	2.47

Table 3. Total phenolics (mg GAE/g DW) and percentage of total carbohydrates in rooting zone tissues of *Dracaena marginata* Lam. rooted air-layer as affected by different combinations of IBA, phloroglucinol (PG) and seaweed extract (SWE) during 2018 and 2019 seasons.

IBA and PG used concentrations were 300 ppm for each, while SWE rate was at 2 ml SW/ L. Each air layer received 10 ml of the corresponding material, and control treatment received 10 ml distilled water.

least rooting percentage supports the idea that the phenolics induced by these treatments are inhibitory to rooting. Consistently, it has been demonstrated that polyphenol compounds act as rooting cofactors whilst other phenolic compounds have a negative impact on adventitious root formation (Osterc et al., 2004). Urbanek Krajnc et al. (2012) found that SWE elicits high the total phenolics in leaves and enhances growth of pelargonium cuttings. Similar results were reported by Jacygrad and Pacholczak (2010) in ninebark cuttings sprayed with SWE. Conversely, Basak et al. (1995) recorded a reduction in polyphenolics in cuttings of certain ornamental plants in response to exogenous auxin application.

However, total carbohydrates percentage exhibited considerably higher values in tissues of treated air-layers than that of the control. Carbohydrates percentage in tissues of air-layers treated with SWE alone or in combination with PG had no significant differences with the control untreated ones, as the three of which contained the lowest values. The other treatments slightly varied from each other. These results are correspondent those to obtained by Kozlowski and Pallardy (1997). Agulló-Antón et al. (2011) observed over a twofold increase in soluble sugars in cuttings of Dianthus caryophyllus treated with auxins. The exogenous auxin application stimulates the mobilization of carbohydrates in leaves upper stem and and the increases translocation of assimilates towards the rooting zone (Agulló-Antón et al., 2011). A significant rise in total soluble sugars after the application of SWE was reported by Sivasankari et al. (2006) and Rathore et al. (2009). According to Khan et al. (2009), SWE causes an increase in cuttings' leaves content of chlorophyll and stimulates gas exchange, and accordingly photosynthetic rate. This directly affects the accumulation of carbohydrates, which is crucial for successful rooting in vegetatively propagated plants (Costa et al., 2007). It has repeatedly been shown that sugar levels within the tissues of cuttings or air-layers are

positively-correlated with adventitious roots formation (Ahkami *et al.*, 2009). Some authors found poor correlation between carbohydrate contents of cuttings and rooting capacity. In this context, Kozlowski and Pallardy (1997) reported that there is a threshold level of carbohydrate necessary for rooting below which development of roots stops. Once the minimal level is available, energy required throughout the rooting period is adequately supplied resulting in successful rooting of cuttings and air-layers.

Correlation study:

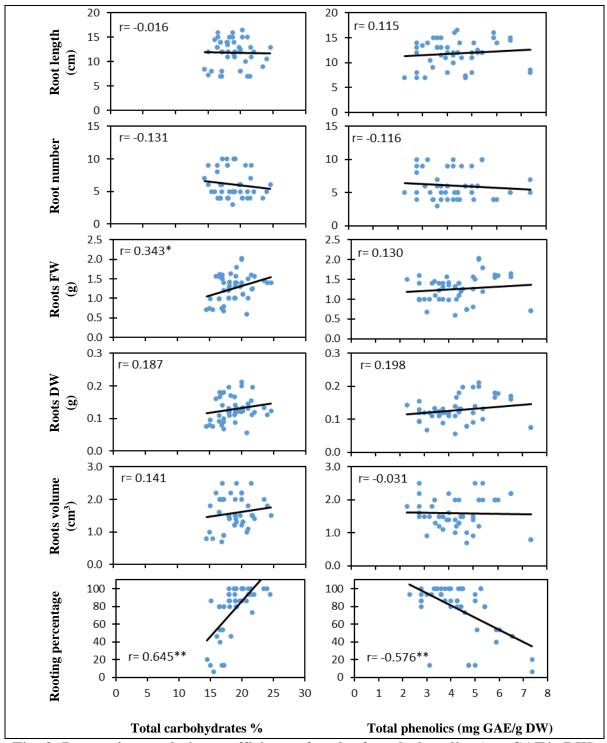
Pearson's correlation coefficients between the internal levels of total phenolics and total carbohydrates in root zones tissues of air layers and all rooting characteristics of air-layers under the effect of different combinations of IBA, PG and SWE were recorded in Fig. (3). Rooting percentage exhibited high significant positive interrelationship with total carbohydrates While, percentage (r= 0.645). high significant negative correlation (r = -0.576)was found between the internal contents of total phenolics in root zone tissues and rooting percentage. At the same time, there were no significant relations between phenolics and carbohydrates contents within root zone tissues and the other studied characteristics.

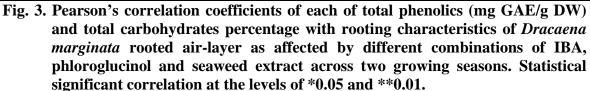
CONCLUSION

Increasing the efficiency of the air layering protocol for D. marginata was possible employing phloroglucinol and seaweed extract in combination with IBA. rooted air-layers Percentage of was pronouncedly increased from 11.11% in the untreated air-layers to reach 46.67 to 100% in the treated ones. Considering the traits of the emerging roots, IBA alone or in combination with PG and SWE exhibited the best results. Improved rooting of air-layers was found to be correlated with higher concentration of carbohydrates and low concentration of phenolics. It is therefore recommended to pretreat air-layers of D. marginata with a mix of IBA, PG and SWE to attain higher rooting percentage and better root traits.

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تطوير ممارسات الترقيد الهوائي لإكثار نبات الدراسينا .Dracaena marginata Lam باستخدام الفلوروجلوسينول ومستخلص الأعشاب البحرية كمساعدات أو بدائل لإندول حمض البيوتريك

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في دراسة موزعة بنظام القطاعات كاملة العشوائية، تم تقييم تأثير ثماني معاملات تجريبية تمثل جميع التوافيق الممكنة بين حمض البيوتريك في صورة ملح بوتاسيوم (٣٠٠ جزء بالمليون) والفلور وجلوسينول (٣٠٠ جزء بالمليون) ومستخلص الطحالب البحرية (٢٨ل/لتر) على تجذير التراقيد الهوائية لنبات الدراسينا. أجريت الدراسة تحت ظروف الصوبة الخشبية بمشتل نباتات الزينة بكلية الزراعة، جامعة أسيوط، مصر خلال موسمي نمو متتابعين ٢٠١٨ و ٢٠١٩. أظهرت جميع التراقيد الهوائية التي تلقت معاملات منفردة أو مشتركة من إندول حمض البيوتريك، الفلور وجلوسينول (٢٠٠ و ٢٠١٩. ومستخلص الطحالب زيادات معنوية في نسبة التجذير مقارنة بالكنترول. وتم تسجيل أعلى نسب التجذير عند استخدام معاملة الفردية من الفلور وجلوسينول أو المعاملة المشتركة من حمض البيوتريك مع مستخلص الطحالب أو المواد الثلاثة مجتمعة بدون فروق معنوية بينها. كما أن التراقيد المعاملة بحمض البيوتريك مع مستخلص الطحالب أو المواد الثلاثة مجتمعة بدون فروق معنوية بينها. كما أن التراقيد المعاملة بحمض البيوتريك مع مستخلص الطحالب أو المواد الثلاثة وتوازن بين الحج والوزن. كما لوحظ ار تباط موجب بين التركيزات المرتفعة للكربو هيدرات الكلية وكل من وتوازن بين الحجم والوزن. كما لوحظ ار تباط موجب بين التركيز ات المرتفعة للكربو هيدرات الكلية وكل من ووزن وحجم الجذور. بينما كان الارتباط سالبا بين تركيز الفينولات المرتفعة للكربو هيدرات الكلية وكل من ووزن وحجم الجذور. بينما كان الارتباط سالبا بين تركيز الفينولات المرتفعة للكربو هيدرات الكلية وكل من حين لم تظهر أي علاقة معنوية بين المواصفات الجذرية الناجمة ومحتوى منائية وكل من نسبة التجذير من كل من حين لم تظهر أي علاقة معنوية بين المواصفات الجذرية الناجمة ومحتوى منطقة التجذير من كل من الكربو هيدرات والفينولات.