

Factors Affecting Adventitious Root Formation and Subsequent Growth and Flowering of *Tabernaemontana divarticata*

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Abstract:

Tabernaemontana divarticata cuttings were dipped in talk powder contained IAA, IBA and NAA, each used individually at 1000ppm, then planted in different rooting media; clay, peat moss, peat + perlite (1:1 v/v), peat + sand (1:1) and vermiculite to define the optimum combinations of rooting media and auxin for stimulating root formation, shoot growth and flowering. Cuttings showed considerable responses to growth regulators compared to control, but IAA and IBA were more pronounced than NAA. The mixture of peat + perlite resulted in the highest rooting percentage followed by peat moss. The high rootability was associated with high C/N ratio in cutting tissues. The combined treatment of either IAA or IBA with peat + perlite gave the maximum rooting percentage and produced the best growth and flowering characteristics. Thus, it could be recommended to use this combination for commercial propagation of *Tabernaemontana divarticata* by cuttings.

Keywords: Growth regulators, rooting media, cutting, *Tabernaemontana divarticata*.

Introduction

Crape jasmine (*Tabernaemontana divarticata*) belongs to Apocynaceae family (8 feet high and 6 feet wide) is a pretty little shrub with a rounded shape flowers. And look like rounded mounds of shiny green leaves. The shrubs flower in spring offering generous amounts of white fragrant blossoms.

Tabernaemontana divarticata cuttings difficult-to-root are characterized by low success and slow growth of the new generated plants. Rooting medium and auxin are considered the most effective factors that exert a strong stimulating influence on the rooting capacity of cuttings, development of adventitious roots, growth of shoots and flowering.

Some investigators declared the favourable effects of auxins on rootability of many ornamental shrubs, El-

Sallami and Mahros (2000) reported that the rooting percentage of poinsettia cuttings was increased after soaking in 50 ppm solutions of IAA, IBA and NAA for 20 hours. Tripathi *et al.* (2003) observed that *Euphorbia pulcherrima* cuttings treated with NAA and IAA, each at 2000 ppm resulted in the highest rooting percentages. Shakouri *et al.* (2012) demonstrated that NAA stimulated the root system of *Dracaena sanderiana* cuttings, Rahdari *et al.* (2014) found that treating cuttings of *Cordyline terminalis* with NAA at 2000 ppm + IBA at 1000 ppm in combination increased the rootability and improved root characteristics of the new produced plants. Ramtin *et al.* (2011) showed that treating poinsettia cuttings with IBA at 1000 ppm increased root length and improved plant characteristics. Saffari and Saffari (2012) re-

ported that treated *Dodonaea viscosa*, L. cuttings with 4000 ppm IBA increased rooting percentage and number of roots. Fouda *et al.* (2012) found that both of IBA at 2000 ppm and NAA at 1000 ppm increased rooting percentage, number and root length of lemon verbena cuttings.

Concerning the effect of rooting media on root formation, vegetative growth and flowering, Bosila *et al.* (2010) cleared that *Bougainvillea glabra* cuttings planted on peat moss medium gave the highest rooting percentage, number of roots per cutting and root length. Hassanein (2013) reported that growing *Ficus hawaii* cuttings on peat moss showed the highest rooting percentage while, the lowest rooting percentage were obtained with perlite medium. El-Maadawy *et al.* (2011) found that using the mixture of peat moss + clay (2:1) and peat moss + sand + clay (2:1:1) had a favorable effect on increasing growth parameters of schefflera plants. They added that plants grown on the mixture of peat moss + sand (1:1) or (2:1) decreased vegetative growth parameters. Similar results were obtained by, Sardoei and Rahbarian (2014) on *Ficus benjamina* plants.

The aim of this study was to define the most suitable synthetic root-promoting growth regulator (auxin) and rooting medium for more successful propagation by cuttings, vegetative growth and flowering of *Tabernaemontana divarticata* plants.

Materials and Methods

This work was carried out at the Experimental Farm of Assiut University during 2013-2014 and 2014-2015 seasons to verify the best growth regulator (auxins) and rooting media

for commercial production of *Tabernaemontana divarticata* plants. by cuttings.

In August of both seasons, tip cuttings (10-15 cm long and 0.3-0.5 cm thickness) were taken from healthy stock plants. Cutting bases were dipped in the mixtures of talk powder with indole acetic acid (IAA), indole butyric acid (IBA), or naphthalene acetic acid (NAA), each used individually at a concentration of 1000ppm, while the control cuttings were dipped in talk powder alone. Cuttings of each treatment were planted in 20 cm diameter pots (10 cuttings/pot) filled with different rooting media; clay, vermiculite, peat moss, peat moss + perlite (1:1 v/v) or peat moss + sand (1:1 v/v). The experiment had 20 treatments (5 rooting media x 4 growth regulators) consisting of a split plot design rooting media as main -plot and growth regulators as sub -plot. The treatments replicated 4 times, each experimental unit contained 30 cuttings. The pots were placed under greenhouse conditions with ambient temperatures of 20-25°C and covered by tightly polyethylene film to maintain high relative humidity.

The percentage of rooted cuttings were recorded after 8 weeks from planting, then three cuttings bases of each treatment were sampled and dried (70°C for 48h) to determine total carbohydrates contents according to Hansen and Moller (1975) and total nitrogen content by semi-micro Kjeldahl method according to Black *et al.* (1965). The ratio of carbohydrates: nitrogen (C/N) was calculated as an indicator of rooting capability.

The rest of new plants were tip pinched and transplanted singly into 25-cm plastic pots filled with the same potting media to follow up the growth rate and flowering of different treatments. Data of shoot, root and flowering characteristics of the newly produced plants were recorded in July of both seasons. Data obtained were statistically analyzed according to Snedecor and Cochran (1989).

Results and Discussion

Rooting percentage

The percentage of rooted cuttings of *Tabernaemontana divarticata* was significantly increased by using the different growth regulators compared to control during the two seasons (Table 1). However, the maximum rooting percentages were obtained from both IAA and IBA showing similar promotive effect, giving 78.4 and 81.2% for IAA, as well as 75.8 and 78.2% for IBA in the first and second season, respectively. These results are in harmony with those found on *Ficus* spp. by Sundaram and Rangaswamy (1994), Mahros *et al.* (1994) and El-Nashar (2000). They reported that auxin application to stem cuttings stimulates the rooting and increased adventitious root formation in their bases as a result of an increase in the cambial activity, root initial formation, and primordia differentiation and elongation.

In the meantime, peat moss, peat + perlite, peat + sand, and vermiculite media increased the percentage of rooted cuttings compared to clay medium. However, the mixture of peat + perlite was the most effective on increasing the rootability resulting in the highest percentages, 83.0 and 84.7% followed by peat moss alone giving 76.9 and 77.9% for the first and second season, respectively. These results are in agreement with those reported by Abo-Hassan *et al.* (1994) on *Ficus infectoria* cuttings.

The favourable effect of peat moss and peat + perlite media on the rooting may be due to maintain high moisture for the cuttings and permit penetration of air to their bases. In this connection, Hartmann *et al.* (2002) stated that an ideal rooting medium provides sufficient porosity to allow good aeration, has a high water-holding capacity and yet is well drained.

The interaction showed that the treated cuttings were treated with either IAA or IBA and grown on the mixture of peat + perlite resulted in the maximum rooting percentage, were 92.5 and 97.9% for IAA, and for IBA they were 94.7 and 96.9% in the first and second season, respectively.

Table 1. Percentage of rooted cuttings (%) in *Tabernaemontana divarticata* as affected by rooting media and growth regulators during 2013/2014 and 2014/2015 seasons.

Media (M)	Growth regulators (G.R.)									
	First season					Second season				
	Control	IAA	IBA	NAA	Mean	Control	IAA	IBA	NAA	Mean
Clay	30.2	63.5	54.6	43.5	48.0	36.9	60.1	59.1	48.0	51.0
Peat moss	56.8	83.5	88.0	79.1	76.9	63.0	88.2	84.5	75.7	77.9
Peat+Perlite	61.3	92.5	94.7	83.5	83.0	65.8	97.9	96.9	78.0	84.7
Peat + sand	56.0	76.9	68.0	70.2	67.8	59.1	84.0	78.3	71.8	73.3
Vermiculite	36.9	75.6	73.5	56.8	60.7	45.8	77.0	73.0	63.5	64.8
Mean	48.2	78.4	75.8	66.6		53.9	81.2	78.2	67.2	
L.S.D.0.05	M:0.8		GR:0.5		MxGR:1.1	M:0.7		GR:0.6		MxGR:1.4

C/N ratio in cutting tissue

Data in Table (2) indicated that rooting behaviour of different treatments showed logical relationship to C/N ratio in cutting base. It is noticeable that application of IAA and IBA which gave the best rootability resulted in the highest values of C/N ratio. Similar results were obtained by Barham *et al.* (2000) on *Ficus retusa* and *Ficus variegata*, and Mahros (2000) on *Bougainvillea* spp. Besides, the high rootability of cuttings grown in the mixture of peat + perlite

was associated also with high C/N ratio. Smalley *et al.* (1991) demonstrated that carbohydrates and nitrogen compounds are capable of stimulating root formation, hence rooting potential has been linked to them. The low nitrogen-high carbohydrates balance (high C/N ratio) in cuttings, which in many cases seems to favour rooting. Mahros (2000) found that carbohydrates content was higher in easy-to-root than in difficult-to-root cuttings of bougainvillea.

Table 2. C/N ratio in tissues of *Tabernaemontana divarticata*. cuttings as affected by rooting media and growth regulators during 2013/2014 and 2014/2015 seasons.

Media (M)	Growth regulators (G.R.)									
	2013-2014 season					2014-2015 season				
	Control	IAA	IBA	NAA	Mean	Control	IAA	IBA	NAA	Mean
Clay	7.5	10.4	10.2	8.7	9.2	8.0	10.9	10.3	8.9	9.5
Peat moss	9.6	13.8	12.8	11.0	11.8	9.1	14.0	13.6	10.9	11.9
Peat+Perlite	10.5	15.8	14.2	13.0	13.4	10.8	15.6	14.7	13.4	13.6
Peat+sand	9.0	11.7	11.8	10.1	10.7	8.8	11.3	11.2	10.2	10.4
Vermiculite	9.1	11.4	11.6	10.0	10.5	8.8	12.0	11.7	10.0	10.6
Mean	9.1	12.6	12.1	10.6		9.1	12.8	12.3	10.7	
L.S.D.0.05	M:0.4		G.R:0.2		G.RxM:0.5	M:0.4		G.R:0.3		MxGR:0.6

Vegetative and root characteristics

It is interesting to note a positive relationship between the highest rooting percentage and the best growth and quality after transplanting. Application of IAA showed the tallest plants, thickest stem and highest number of branches and leaves. Meanwhile, IBA gave the heaviest

fresh and dry weights of aerial parts and roots. However, NAA resulted in some increments in such parameters compared to control (Tables 3 & 4). These increases in vegetative parameters could be attributed to the important role of auxin in plant growth. Noggle and Fritz (1989) demonstrated that the increase in plant

height as a result of IAA application at a suitable level due to enhance cell division and/or cell elongation. Besides, the stimulatory action of auxin in softening the cell wall, increasing its plasticity leading to cell enlargement and consequently stimulating stem length. In addition, Mahros (2000) found that application of IBA to bougainvillea cuttings increased size of root system, which reflected in producing vigour and healthy shoot system. In this regard, Hartmann *et al.* (2002) stated that the large root size on cutting enhanced shoot growth rate, and root reduction has been associated with shoot stunting. A large size of root system absorbs high rates of water and nutrients which translocate to the shoot system lead to promote its growth and development.

Regarding the effect of rooting media on the vegetative and root characteristics (Tables 3 & 4), it was noticed that the mixture of peat + perlite was the superior medium for improving plant growth parameters as it recorded 33.8 and 34.3 cm for plant height, 9.7 and 10.2 mm for stem diameter, 8.1 and 8.5 for branch number per plant, 18.7 and 17.6 for leaf number, 33.47, 36.51g for fresh weight of the aerial parts, 10.2 and 10.01g for dry weight of the aerial parts, and 10.86 and 9.84g for root fresh weight in the first and second season, respectively. Mostly, peat moss alone achieved the next high values in such parameters. In contrast, vermiculite or peat + sand resulted in the inferior results showing the worst growth.

Several investigators concluded that the rooting medium can affect

the type of root system arising from cuttings and their growth characteristics such as Siraj and Abou-Dahab (1993), Abo-Hassan *et al.* (1994) and Ishtiaq *et al.* (1995 a&b). They declared that cuttings of some species when rooted in sand, produced long, unbranched, coarse and brittle roots, but when rooted in peat moss or a mixture of peat moss + perlite, develop roots that are well branched, slender and flexible, a type much more suited for digging and repotting.

The interaction indicated that the application of IAA or IBA plus peat + perlite medium produced tallest plants, thickest stem, highest number of branches and leaves and heaviest fresh and dry weights of the aerial parts and roots.

Flowering characteristics:

The interactions between rooting media and growth regulators on flower number and weight are illustrated in Figure (1 A & B). It is clear that plants treated with IAA and grown in peatmoss +perlite medium produced the highest number and heaviest flowers.

The best flower characteristics obtained from this treatment could be attributed to the "ideal" root-medium for plant growth as reported by Riviere *et al.* (1990) who pointed out that the mixture of peat + perlite has the most flexible irrigation requirements and is thus the least restrictive in terms of water managements. They added that this medium maintain the balance in the availability of mineral ions as nitrogen, phosphorous and potassium required for growth as a result of various quite specific vascular relationships between the leaves and other parts of the plant; the amount of

photosynthate translocated from leaves is sufficient for flowering processes. In addition, auxin has already been found to play important roles in the endogenous control of flowering. The existence of auxin which regulate the quantity and quality of growth offers interesting possibilities of explaining such developmental events as flowering. A general explanation is on the basis of more carbohydrates synthesis which closely associated with an effect of auxin role on marked increase in leaf formation led to increase in photosyn-

thesis activity, consequently producing more photosynthetic products leading more carbohydrates accumulation. These observations are supported by the findings of Gibson (2005), and Kepinski and Leyser (2005) they concluded that there was a positive correlation among auxin, carbohydrates and metabolism of flowering was found that is very important to produce vigorous plants with dense flowers. The adaptation of flowering dependent on maintaining high carbohydrates.

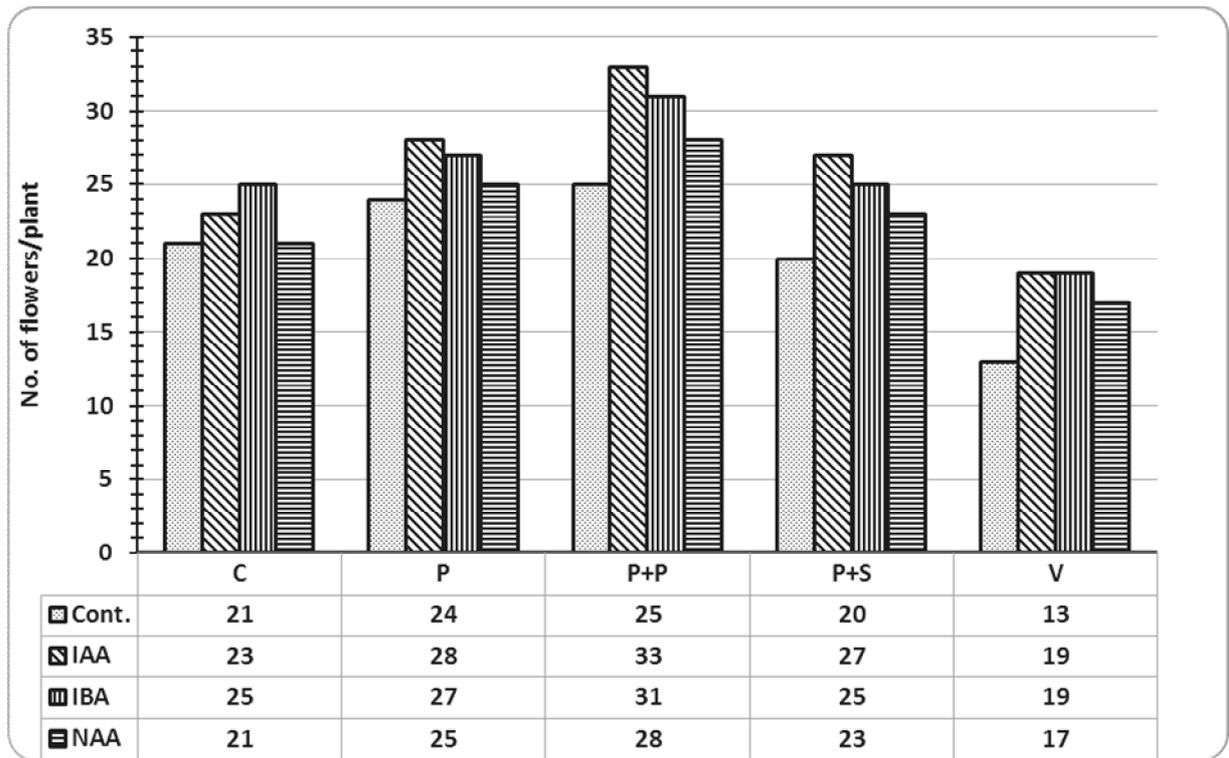
Table 3. Effect of rooting media and growth regulators on vegetative growth characteristics of *Tabernaemontana divarticata* during 2013/2014 and 2014/2015 seasons.

Media (M)	Growth regulators (G.R.)									
	First season					Second season				
	Control	IAA	IBA	NAA	Mean	Control	IAA	IBA	NAA	Mean
	Plant height (cm)									
Clay	22.7	29.0	23.7	24.0	24.8	21.7	30.3	24.7	24.6	25.3
Peat moss	25.7	40.6	30.7	30.0	31.8	27.3	39.7	30.7	29.3	31.8
Peat + Perlite	27.0	46.0	31.3	30.7	33.8	28.7	46.7	32.3	29.6	34.3
Peat + sand	21.3	30.7	27.3	27.3	26.6	23.0	32.3	26.0	26.6	27.0
Vermiculite	21.0	26.7	22.7	21.7	23.0	21.7	28.0	23.7	22.3	23.9
Mean	23.5	34.6	27.1	26.7		24.4	35.4	27.4	26.5	
L.S.D. 5%	M: 1.8		G.R. 1.6		G.R.xM: 3.6	M: 2.0		G.R. 1	G.R. x M: 4.0	
	Stem diameter (mm)									
Clay	7.8	8.6	8.3	8.3	8.2	7.9	8.7	8.5	8.3	8.3
Peat moss	8.6	10.1	9.3	9.1	9.3	9.3	10.4	9.6	9.5	9.7
Peat + Perlite	9.0	10.6	9.6	9.5	9.7	10.1	10.8	10.0	9.8	10.2
Peat + sand	9.2	10.0	9.5	9.0	9.4	9.0	10.1	9.3	9.3	9.4
Vermiculite	8.8	9.6	9.0	8.9	9.1	8.6	9.0	9.3	9.0	9.0
Mean	8.7	9.8	9.1	8.9		9.0	9.8	9.3	9.2	
L.S.D. 5%	M: 0.3		G.R. 0.3		G.R.x M: 0.7	M: 0.3		G.R. 0.3	G.R. x M: 0.7	
	Number of branches/plant									
Clay	5.3	6.0	7.0	6.0	6.0	5.6	7.3	6.6	6.7	6.6
Peat moss	7.3	8.6	7.0	6.6	7.4	7.6	9.3	6.7	7.3	7.7
Peat + Perlite	7.6	9.6	8.3	6.7	8.1	8.3	9.3	8.7	6.6	8.5
Peat + sand	5.3	6.6	6.3	6.6	6.2	5.6	7.0	6.0	7.0	6.4
Vermiculite	5.0	5.5	6.0	6.0	5.6	4.7	7.3	6.3	6.3	6.3
Mean	6.1	7.3	6.9	6.4		6.4	8.0	6.9	6.8	
L.S.D. 5%	G.R. 0.5		M: 0.6		G.R.x M: 1.1	M: 0.7		G.R. 0.6	G.R. x M: 1.3	
	Number of leaves/plant									
Clay	13.9	17.1	17.1	16.9	16.3	13.6	17.2	16.4	16.4	15.9
Peat moss	15.1	19.9	17.9	17.3	17.6	14.4	17.5	17.2	16.5	16.4
Peat + Perlite	16.3	21.8	19.0	17.8	18.7	16.0	19.0	18.4	17.1	17.6
Peat + sand	15.2	17.3	16.9	16.8	16.6	13.0	16.8	16.1	16.1	15.5
Vermiculite	13.4	16.3	16.4	16.0	15.5	13.1	15.9	15.9	15.8	15.2
Mean	14.8	18.7	17.5	17.0		14.0	17.3	16.8	16.4	
L.S.D. 5%	M: 1.5		G.R. 1.3		G.R.xM: 2.9	M: 1.3		G.R. 1.2	G.R. x M: 2.7	

Table 4. Effect of rooting media and growth regulators on fresh and dry weights of aerial parts and roots of *Tabernaemontana divarticata* during 2013/2014 and 2014/2015 seasons.

Media (M)	Growth regulators (G.R.)										
	First season					Second season					
	Control	IAA	IBA	NAA	Mean	Control	IAA	IBA	NAA	Mean	
Fresh weight of aerial parts (g/plant)											
Clay	19.41	33.33	39.03	14.43	26.55	19.84	35.6	38.33	23.2	29.24	
Peat moss	20.47	36.73	39.23	19.67	29.02	21.46	33.2	37.55	25.65	29.47	
Peat + Perlite	23.23	40.53	45.00	25.12	33.47	26.25	42.73	47.95	29.10	36.51	
Peat + sand	15.97	29.00	31.67	17.47	23.60	16.64	25.62	28.13	17.86	22.06	
Vermiculite	14.34	17.33	24.60	13.34	17.50	11.05	19.00	20.38	13.70	16.03	
Mean	18.68	31.38	35.91	18.00		19.05	31.23	34.47	21.90		
L.S.D. 5%	M: 0.49		G.R. 0.71		G.R. x M: 1.59		M: 0.77		G.R. 0.76		G.R. x M: 1.71
Dry weight of aerial parts (g/plant)											
Clay	6.04	10.52	12.59	4.65	8.45	6.07	10.88	11.98	4.13	8.37	
Peat moss	6.38	11.62	12.65	6.12	9.20	7.58	10.88	11.73	4.89	9.00	
Peat + Perlite	4.27	13.82	14.52	8.12	10.2	5.08	13.16	14.98	5.97	10.01	
Peat + sand	4.93	8.87	10.22	5.41	7.36	4.08	7.69	8.79	2.46	6.65	
Vermiculite	4.64	5.59	7.94	4.32	5.63	3.45	5.56	6.37	4.28	5.30	
Mean	5.25	10.08	11.58	5.72		5.25	9.63	10.77	4.34		
L.S.D. 5%	M: 0.16		G.R. 0.23		G.R. x M: 0.51		M: 0.24		G.R. 0.23		G.R. x M: 0.54
Fresh weight of roots (g/plant)											
Clay	9.90	8.18	10.78	10.10	9.74	6.50	8.45	10.8	7.58	8.33	
Peat moss	6.40	8.83	10.43	6.03	7.93	6.88	8.38	10.53	9.67	8.87	
Peat + Perlite	5.84	15.64	16.08	5.88	10.86	5.48	13.28	15.33	5.26	9.84	
Peat + sand	2.34	7.83	9.95	6.48	6.65	2.35	7.58	8.93	7.48	6.59	
Vermiculite	5.45	6.10	6.68	5.50	5.94	4.73	4.75	5.49	5.43	5.10	
Mean	5.99	9.32	10.78	6.80		5.19	8.49	10.22	7.08		
L.S.D. 5%	M: 0.37		G.R. 0.49		G.R. x M: 1.09		M: 0.51		G.R. 0.36		G.R. x M: 0.79
Dry weight of roots (g/plant)											
Clay	4.50	3.72	4.89	4.59	4.43	2.83	3.67	4.69	3.29	3.62	
Peat moss	2.91	4.01	4.74	2.74	3.60	2.99	3.64	4.58	4.20	3.85	
Peat + Perlite	2.65	6.93	7.31	2.67	4.89	2.38	5.73	6.66	2.29	4.27	
Peat + sand	1.06	3.65	4.52	2.94	3.10	1.02	2.98	3.88	3.25	2.78	
Vermiculite	2.48	2.77	3.03	2.50	2.70	2.05	2.06	2.39	2.36	2.22	
Mean	2.72	4.22	4.90	3.09		2.25	3.62	4.44	3.08		
L.S.D. 5%	M: 0.022		G.R. 0.17		G.R. x M: 0.49		M: 0.22		G.R. 0.15		G.R. x M: 0.35

A:



B:

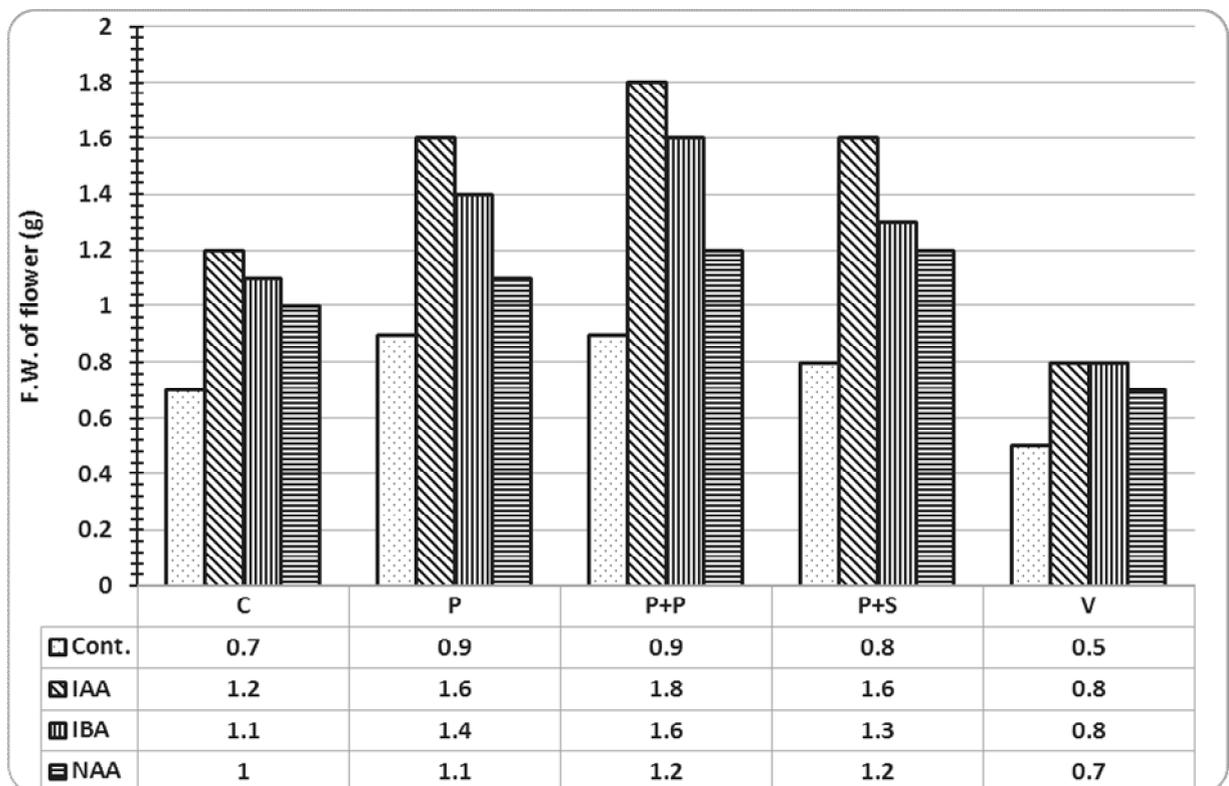


Fig (1): Effect of rooting media and growth regulators on flower number (A) and fresh weight (B) of *Tabernaemontana divarticata* as an average of two seasons.
 (Clay=c, Peatmoss = P, Peatmoss + perlite =P+P, Peatmoss +Sand = P+S, Vermiculite = V)

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عوامل مؤثرة على تكوين الجذور العرضية ونمو ازهار التبرنامونتانا ديفارتيكاتا

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الملخص

أجريت هذه الدراسة بمزرعة أبحاث نباتات الزينة بكلية الزراعة - جامعة أسيوط خلال الموسمين ٢٠١٣ / ٢٠١٤ و ٢٠١٤ / ٢٠١٥ لتحديد المعاملة الأكثر تأثيراً في زيادة قدرة عقل التبرنامونتانا على التجذير وتنشيط النمو الخضري والجذري والزهري. شملت الدراسة بعض منظمات النمو (اندول حمض الخليك ، إندول حمض البيوتريك ، نفتالين حمض الخليك، كل على حده بتركيز ١٠٠٠ جزء في المليون مخلوطاً بمسحوق التلك) وبيئات التجذير: طمي ، فيرميكوليت ، بيت موس ، بيت موس + بيرليت (١:١) ، بيت موس + رمل (١:١) حيث غمست قواعد العقل في خليط الهرمون وزرعت بعد ذلك مباشرة في تلك البيئات.

وكانت أهم النتائج ما يلي:

- أدى استعمال إندول حمض الخليك أو اندول حمض البيوتريك إلى زيادة قدرة العقل على التجذير حيث أعطيا أعلى نسبة تجذير. بينما أظهر نفتالين حمض الخليك تأثيراً أقل منهما.
- أعطت العقل المنزرعة في مخلوط البيئة المكونة من البيت موس + بيرليت أعلى نسبة تجذير تلاها في ذلك بيئة البيتموس.
- ارتبطت القدرة العالية لتجذير العقلة بزيادة محتوى قاعدتها من نسبة الكربوهيدرات : النيتروجين بأنسجتها.
- أظهرت المعاملة المشتركة المكونة من إندول حمض الخليك أو إندول حمض البيوتريك مع بيئة البيتموس + بيرليت أعلى نسبة تجذير كما حسنت مواصفات النمو الخضري والجذرية والزهريّة. ولذا يمكن النصح باستعمالها عند اكثار نبات التبرنامونتانا بالعقل للإنتاج التجاري.