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Influence of Some Plant Growth Substances on Shoot and Root Initiations of Chrysanthemum Explants *in Vitro*.

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ABSTRACT

The present research was carried out to study the effect of some growth regulators such as 6- benzyl amino purine (BAP) at 0.5, 1.0, 2.0 and 5.0 mg/L, Naphthalene acetic acid (NAA) at 0.1 and 0.5 mg/L and their combinations in the initiation media. Besides studding the influence of such growth regulators (BAP and NAA) at concentration of 0.2, 1.0, 2.0 or 4.0 mg/L BAP and 0.1 or 0.2 mg/L NAA in the multiplication media. In addition, different auxins {i.e., Indole-3-butyric acid (IBA), Naphthalene acetic acid (NAA) and Indole-3 acetic acid (IAA)} at 0.1, 0.3, 0.5 and 1.0 mg/L were applied separately in the rooting media on the *in vitro* micropropagation and acclimatization of *Dendranthema grandiflorum* plants**.** The chrysanthemum cultivar used in this research was namely flyer. To evaluate the adaptation degree of chrysanthemum cultivar to the culture medium composition, the single nodal explant (0.5 – 1.0 cm) were surface sterilized and cultured on MS medium supplemented with sucrose (30 g/L), agar (7 g/L). The initiated shoots were excised and cultured on the multiplication and rooting media. The obtained results indicated that explants grown on medium containing 1.0 mg/L BAP + 0.5 mg/L NAA produced the highest shoots number. While, explants grown on medium containing 2.0 mg/L BAP + 0.1 mg/L NAA produced the tallest shoots and internodes. Moreover, explants grown on medium fortified with 1.0 mg/L BAP + 0.1 mg/L NAA produced the highest leaves number. The obtained results also indicated that, the explants grown on medium supplemented with 1.0 mg/LBAP+ 0.2mg/L NAA produced the highest shoots number. While, those grown on the multiplication medium containing 1.0 mg/L BAP + 0.1 mg/L NAA produced the longest shoots and internodes. Finally, shoot tips grown on medium containing 2.0 mg/l BAP + 0.1 mg/L NAA formed the highest leaves number. Moreover, rooting of the multiplied shoots were achieved on the same medium used in the multiplication stage except for the macro-elements of MS medium which used at half strength, sucrose at 20 g/L and 0.5 mg/L IBA which formed the roots initiation within a few days, followed by the medium which contained1.0 mg/L IBA which produced the highest roots number. While, medium containing 0.1 mg/L IBA resulted in the tallest roots per explants. For the acclimatization process, the combination of peat moss + perlite + vermiculite (1:1:1 v/v) showed its superiority over all the other growing media used, as it gave 100% survival percentage (%), increase in leaves number per shoots, shoots length (cm), and internodes length.

INTRODUCTION

Development of procedures for rapid *in vitro* clonal propagation of chrysanthemum may be of great commercial value to the chrysanthemum industry. Tissue culture techniques should minimize the time necessary for production of new cultivars into the commercial market and thus increase the availability of chrysanthemum plants with improved horticultural characteristics. These techniques may be also of potential benefit for the mass propagation of existing commercial cultivars. After the first works of Ben-jaacov and Langhans (1972) on the *in vitro* development and rooting of chrysanthemum shoot tips, many studies were tried to find efficient methods to propagate lines of commercial varieties as mentioned by Gartsson and Andersson (1985). In the *in vitro* propagation process, it is important to establish which factors control the mechanism of shoot multiplication, root initiation on cultured shoot tips and which are for the successful transfer of plantlets obtained *in vitro* to soil. Some of these factors have been extensively studied: cultures, media factors (e. g. nutritional or hormonal) and environmental factors. However, their functions can not be generalied to all plant species.

In recent years, plant cell and tissue culture techniques have developed into very powerful tools for the propagation of ornamental species. The micropropagation of plants through tissue cultures in general can provide unlimited number of plantlets from a single explant. In addition, this technique could eliminate virus's growth with the regenerated plantlets, rapid clonal multiplication of valuable specimens, vegetative propagation of species that are difficult to propagate and propagation of clones all over the year.

Successful application of micropropagation through tissue culturing of chrysanthemum genotypes is not general. It is problematic in case of some cultivars, in the 4 stage: Initiation, multiplication, rooting *in vitro* and acclimatization under greenhouse conditions. Accordingly, the objective of this study were to examine the effect of some cytokinins and auxins at different concentrations on shoots multiplication and rooting of shoot tips as well as plantlets acclimatization under greenhouse conditions.

MATERIALS AND METHODS

The *Dendranthema grandiflorum* Ram. (Fam: *Asteraceae*) cultivar of white cv. Flyer was examined in this study. The single nodal explant (0.5 – 1.0 cm length) were taken from the vegetative plants grown in the greenhouse. The apices of vegetative plants were removed and four weeks later, the new axillary shoots were cut off during the month of January (2014) from plants grown in the Nursery of Agriculture college, Mansura University. After removing the leaves longer than 5.0 mm, the shoots were cut into single nodal segment with one axillary bud. The work was conducted during the period from 2014 to 2016 at the Tissue culture laboratories of vegetable and Floriculture Department, Faculty of Agriculture, Mansoura university. Aseptic cultures established from the nodal segments were surface sterilized in three steps : rinsed

in a distilled water for 10 min., immersion in sodium hypochlorite at 2.25% containing few drops of Tween – 20 wetting agent for 20 min. and rinsing 4 time in a sterile distilled water changed for 2 min. each, the study included the following 4 tests :

The first test was concerned with culturing single nodal exptants on the initiation medium described by Murashige and Skoog (1962) medium supplemented with sucrose (30g/L), agar (7.0 g/L), 6-benzylamino purine (BAP) at 0.5 , 1.0 , 2.0 , and 5.0 mg/L and naphthalene acetic acid (NAA) at 0.1 and 0.5 mg/L.

The second test was carried out on the sterile shoot tips (15 – 20 mm length) obtained from proliferated shoots and transferred onto multiplication media consisted of the mineral salt formulation and vitamins as described by MS, sucrose (30.0g/l), agar (7.0g/L) and 6-benzylamino purine (BAP) at concentrations of 0.2, 1.0, 2.0 and 4.0mg/L in the presence of NAA at 0.1 or 0.2 mg/L.

The third test was concerned with evaluating the effect of IAA, IBA and NAA at different concentrations on the rooting and survival percentage of plantlets. Whereas, the shoot tip explants (15-20 mm length) multiplied in the multiplication media were transferred on rooting medium containing the same medium used in multiplication stage except for the macroelements of MS medium as it used at half strength, sucrose at 20g/L, agar (7.0 g/L) and the different auxins (i. e., IBA, NAA, and IAA) were tested singly at concentrations of 0.1,0.3, 0.5 and 1.0 mg/L .

The last test was designed on the acclimatization test. Healthy and well rooted plantlets of chrysanthemum were taken away from jars. The agar was removed from the plantlets simply by washing with slightly hot tap water (35C^{o}), followed by washing with a commercial fungicide (Rizolex1g/L), and then transferred into plastic pots (5cm) containing one of the following autoclaved media; loamy soil + Peat moss + vermiculite (1:1:1v/v), loamy soil + peat moss + sand (1:1:1v/v) or peat moss + perlite + vermiculite (1:1:1 v/v). Pots with plantlets were transferred to glass containers (100*60*25 cm) existed in a glass room. Tab water was added to the bottom of the glass containers as a thin, layer (0.5 cm) with adding the salts of MS medium at concentration of ¼ strength. The glass containers were kept covered with a clear polyethylene sheet to maintain high relative humidity and fluorescent light lamp with intensity of 2500 lux in the incubation room for 2 weeks. At the end of the incubation period (2 weeks), the clear polyethylene sheet was completely raised on the glass containers. During the period, the plants were watered with tap water.

The PH was adjusted to 5.8 with NaoH and HCL before agar was added. The culture medium was autoclaved at 120 C^{o} for 20 min.

In all above treatments, medium of 30 ml was dispensed into 200 ml jars for explants culture, one nodal explant was cultured per jar for the initiation stage and one explants were cultured per jar for the multiplication and rooting stage. Cultures were maintained at 23 ± 1C^{o} under a constant fluorescent light (day light F-29) of 2500lux for 16/8 day/night.

Throughout the shoots initiation and multiplication stages of the present study, the following characters were recorded: four weeks later, shoots number (shoot), shoots length, leaves number (leaf)and internodes length (cm). During the root stage, number of days until appearance of visible formed roots was recorded .Two Weeks after incubation the following observations were determined : number of roots per explant, length of roots per explant (cm). and after 4 weeks of acclimatization the survival percentage of the rooted plantlets(%),length of shoots, number of leaves (leaf), and internode length (cm) were recorded.

Statistical analysis

Treatments were arranged in a complete randomized design (CRD) with three replicates each consists of 4 jars.The obtained data were subjected to analysis of variance (ANOVA) by using "Genstat 11.1" (2008). The comparisons between the means were according to Gomez and Gomez, (1984) witha significant level of 5% for all the statistical analyses.

RESULTS AND DISCUSSION

Initiation stage.

1:Effect of BAP, NAA and their interactions on shoots number and shoots length(cm)of *Dendranthema grandiflorum* explants after incubation for 4 weeks on initiation medium.

Data in Table (1) indicated that, explants grown on medium containing 1.0 mg/L BAP produced significantly the highest numbers of shoots (2,83 shoot). In addition, number of shoots was increased with adding NAA at 0.5 mg/L in the initiation medium. Concerning the interaction effects, it was evident from the same Table that, explants grown on a proliferation medium supplemented with 1.0 mg/L BAP + 0.5 mg/L NAA produced the highest shoot number (3.67 shoots for). While, medium containing 0.5 mg/l BAP + 0.1 mg/L NAA produced the lowest number of shoots (1.0 shoot).This was in agreement with Ali *et al*., (2007) who recommended that 1.0 mg/l BAP was the most optimum BAP concentration for the regeneration of plantlets and any increase or decrease in its concentration caused a decrease in multiplication rates. Data presented in the same Table showed that, explants grown on medium fortified with 2.0 mg/L BAP formed the tallest shoots (4.67 cm). Moreover, explants grown on medium supplemented with 0.5 mg/L NAA formed the tallest shoots (4.14 cm) than those grown on medium containing 0.1 mg/L NAA. Concerning the interaction effect, it was obvious from the same Table that, explants grown on medium containing 2.0 mg/L BAP + 0.1 mg/L NAA formed the tallest shoots (4.93 cm) than those grown on the other media. However, the explants grown on medium containing 5.0 mg/L BAP+ 0.1 mg/ L NAA produced the shorter shoots (3.60 cm).

This could refer to the mode of action of the plant growth regulators, as it stimulate the growth and division of the plant cell in the lower concentration or in the optimal concentration, but in the excess concentrations it inhibit the plant cell growth and could be a murderer.

Table 1. Effect of BAP, NAA and their interactions on shoots number (shoot) and shoots length (cm) of *Dendranthema grandiflorum* explants after incubation for 4 weeks on initiation media.

Treatments	Measured data (Average) Shoots					
	Number (shoot)			Length (cm)		
Conc. Of BAP mg/L	Conc. Of NAA mg/L					
	0.1	0.5	Mean of (A)	0.1	0.5	Mean of (A)
0.5	1.00	3.65	2.33	3.47	4.23	3.85
1	2.00	3.67	2.83	4.13	4.03	4.08
2	1.33	3.00	2.17	4.93	4.40	4.67
5	2.33	3.00	2.67	3.60	3.90	3.75
Mean of (B)	1.67	3.33		4.03	4.14	
L.S.D (5%)	(A)	(B)	(AB)	(A)	(B)	(AB)
	0.90	0.62	1.27	2.99	2.12	4.23

2: Effect of BAP, NAA and their interactions on leaves number (leaf) and internodes length (cm) of *Dendranthema grandiflorum* explants after incubation for 4 weeks on initiation medium.

Data described in Table (2) revealed that, the explants grown on medium supplemented with 1.0 mg/L BAP formed the greatest number of leaves per explants (12,83 leaves). While, explants grown on medium containing 0.5 mg/L NAA produced the highest number of leaves per explants (11.25 leaves) than those grown on medium containing the lower concentration of NAA. Data presented in the same Table showed that using a medium fortified with 1.0 mg/L BAP + 0.1 mg/L NAA produced significantly the higher leaf numbers per explant (14.00 leaves). While, the lowest leaves number (6.00 leaves) was produced with growing on medium containing 0.5 mg/L BAP + 0.1 mg/L NAA. The results tabulated in the same Table cleared that, the tallest terminal three internodes (0.47 cm for each) were formed with explants grown on medium containing 1.0 mg/L BAP and 2.0 mg/L BAP, respectively. Moreover, the longest terminal three internodes (0.48 cm) resulted from shoot explants grown on medium supplemented with 0.1 mg/L NAA. Concerning the interaction effect, it was evident from the same Table that, the tallest terminal three internodes (0.53 cm) were obtained with shoot explants grown on medium containing 2.0 mg/L BAP + 0.1 mg/L NAA comparing with the other interactions.

Table 2. Effect of BAP and NAA on leaves number (leaf) and internodes length (cm) of *Dendranthema grandiflorum* after incubation for 4 weeks on the initiation media.

Treatments	Measured data (Average) Leaves number (leaf)			Internodes length (cm)		
Conc. of	Conc. of NAA mg/L					
BAP mg/L	0.1	0.5	Mean (A)	0.1	0.5	Mean (A)
0.5	6.00	11.00	8.50	0.43	0.30	0.37
1	14.00	11.67	12.83	0.50	0.43	0.47
2	10.33	11.67	11.00	0.53	0.40	0.47
5	11.00	10.67	10.83	0.43	0.47	0.45
Mean of (B)	10.33	11.25		0.48	0.40	
L.S.D (5%)	(A)	(B)	(AB)	(A)	(B)	(AB)
	4.04	2.68	5.72	0.40	0.28	0.56

Multiplication stage.

3: Effect of BAP, NAA and their interactions on, shoots number (shoot) and shoots length (cm) of *Dendranthema grandiflorum* explants after incubation for 4 weeks on *multiplication medium.*

Date in Table (3) showed that, explants grown on a multiplication medium supplemented with BAP at 1.0 or 4.0 mg/L produced significantly the highest number of shoots (3.33 shoot for each). While medium containing the lower concentration of BAP (0.2 mg/L) produced the weakest shoot numbers of 1.67 shoot. In addition, as for effect of NAA concentrations in this parameter, it was clear that using medium received the higher concentration of 0.2 mg/L, produced the highest shoot numbers of 3.00 shoots. This was in agreement with Khan *et al*., (1994) who found that each increase in BAP concentration, followed by increase in shoot the proliferation %. Concerning the interaction effect, it was evident from the same Table (4) that, the explants which grown on a multiplication medium containing 1.0 mg/L BAP + 0.2 mg/l NAA and 2.0 mg/L BAP + 0.2 mg/L NAA produced the highest number of shoots (3.67 shoots for each) than the other interactions. Data reported in the same table revealed that, explants growing on medium containing 1.0 mg/L BAP, produced the significant tallest shoots of 1.82 cm. While, those grown on a medium containing 4.0 mg/L BAP formed the shorter shoots (1.35cm). Moreover, explants cultured on medium fortified with 0.1 mg/L NAA produced the tallest shoots (1.63 cm), while in contrary the shoot explants grown on medium containing the higher concentration of NAA (0.2 mg/L) produced the shortest shoots (1.43 cm). Regarding the interaction effect, it was observed from the same Table that, explants growing on medium containing 1.0 mg/L BAP + 0.1 mg/L NAA produced significantly the tallest shoots (2.20 cm).

4: Effect of BAP, NAA and their interactions on leaves number and internodes length (cm) of *Dendranthema grandiflorum* explants after incubation for 4 weeks on *multiplication medium*.

Data in Table (4) showed that, explants grown on medium supplemented with 0.2 mg/L BAP produced significantly the highest number of leaves per explants (9,00 leaves).The explants cultured on medium containing 0.1 mg/LNAA produced significantly the highest number of leaves (8.92 leaves) than those grown on medium containing 0.2 mg/L NAA (7.75 leaves).

Table 3. Effect of BAP and NAA and their interactions on shoots number (shoot) and shoots length (cm) of *Dendranthema grandiflorum* explants after incubation for 4 weeks on multiplication media.

Treatments	Measured data average Shoots					
	Number (shoot)			Length (cm)		
Conc. of	Conc. Of NAA mg/L					
BAP mg/L	0.1	0.2	Mean of(A)	0.1	0.2	Mean of(A)
0.2	2.00	1.33	1.67	1.53	1.50	1.52
1	3.00	3.76	3.33	2.20	1.43	1.82
2	2.76	3.76	3.17	1.33	1.53	1.43
4	3.33	3.33	3.33	1.43	1.27	1.35
Mean of (B)	2.75	3.00		1.63	1.43	
L.S.D (5%)	(A) 1.48	(B) N.S	(AB) 2.09	(A) 0.40	(B) N.S	(AB) 0.56

Concerning the interaction effect, it was cleared from the same table that, the shoot explants grown on a medium fortified with 0.2 mg/L BAP + 0.1 mg/L NAA produced significantly the highest number of leaves (9.67leaf). While, explants grown on media supplemented with 4.00 mg/L BAP + 0.2 mg/L NAA produced the lowest number of leaves (6.67 leaves). It was clear from data presented in the same table that, the tallest terminal three internodes (0.17 cm) were produced from explants grown on medium containing BAP at 1.0 mg/L. While, shoot explants grown on medium containing 0.1 mg/L NAA produced the tallest terminal three internodes (0.14 cm).

Table 4. Effect of BAP and NAA and their interactions on leaves number (leaf) and internodes length(cm) of *Dendranthema grandiflorum* explants after incubation for 4 weeks on multiplication media.

Treatments	Measured data (Average)					
	Leaves number (leaf)			Internodes length (cm)		
Conc. of	Conc. Of NAA mg/L					
BAP mg/L	0.1	0.2	Mean of (A)	0.1	0.2	Mean of (A)
0.2	9.67	8.33	9.00	0.13	0.11	0.12
1	9.00	7.67	8.33	0.23	0.10	0.17
2	8.67	8.33	8.50	0.11	0.13	0.12
4	8.33	6.67	7.50	0.10	0.07	0.09
Mean of (B)	8.92	7.75		0.14	0.10	
L.S.D (5%)	(A) 1.95	(B) N.S	(AB) 2.76	(A) 0.05	(B) S	(AB) 0.12

Regarding the interaction effect, data reported in the same Table (4) indicated that, explants grown on medium supplemented with 1.0 mg/L BAP + 0.1 mg/L NAA produced significantly the tallest three terminal internodes (0.23 cm) than the other treatments.

Rooting stage.

5. Effect of auxin type and concentrations on number of days until appearance of visible formed roots per explant, roots number (root) and roots length (cm) of *Dendranthema grandiflorum* after incubation for 2 weeks on rooting media.

Data presented in Table (5) showed that, the roots initiation appeared within 6-10 day from culturing on rooting media. The roots which emerged significantly earlier (after 6 days) resulted from the explants grown on the rooting media containing 0.5 mg/L IBA and 0.3 mg/L NAA . While, with explants grown on rooting media containing 0.5 or 1.0 mg/L NAA, the time of emergence of visible root formation was longer (after 10 days for each). Data reported in the same Table indicated that shoot explants grown on rooting medium containing IBA at different concentrations produced the highest number of roots (6.83 roots). While, explant grown on the rooting medium containing IAA produced significantly the lowest number of roots (3.18 root) and formed the shortest roots (0.84 cm). Concerning roots number data reported in the same Table shows the effects of 0.1, 0.3, 0.5 and 1.0 mg/L of auxins. The number of roots per explant increased with increasing IBA or NAA concentrations in the root induction media. The highest roots number were produced when shoot tips were grown on medium containing IBA at 0.5 or 1.0 mg/L significantly recorded the highest values of roots number (9.33 and 10.33 roots) after 2 weeks from the *in vitro* rooting treatments, when compared with all of the other treatments. While the lowest average roots number (1.70 root) after 2 weeks were produced on a medium containing 1.0 mg/L IAA. In the present experiment, it was clear that, the average roots length decreased with increasing of the auxin concentrations.

The medium without hormone induced a significant increase in roots length as compared with shoot tips grown on media containing different concentrations of IBA, NAA, and IAA. The shoot explants grown on medium containing 0.1 mg/L IBA produced the longest roots (2.13 cm), while those grown on a rooting medium containing 1.0 mg/L IAA formed the shortest roots (0.30 cm) after 2 weeks from culturing.

In the present experiment, it was clear that, the average roots length decreased with increase of the auxin concentrations. The medium without hormone induced a significant increase in roots length as compared with shoot tips grown on media containing different concentrations of IBA, NAA, and IAA. The shoot explants grown, on medium containing 0.1 mg/L IBA produced the longest roots (2.13 cm), while those grown on a rooting medium containing 1.0 mg/L IAA formed the shortest roots (0.30 cm) after 2 weeks from culturing. The decrease in roots length was much. with IAA than with IBA and NAA. Similar results were obtained by Rout (2006) who reported according to the result for the species IBA was shown to produce a higher yield of roots compared to the other auxins.

Table 5. Effect of Auxin type IBA, NAA and IAA at different concentrations on number of days until appearance of visible formed roots per explant, roots number (root) and roots length (cm) of *Dendranthema grandiflorum explants* after incubation for 2 weeks on rooting media.

Auxin type	Auxin conc. at mg/L	Growth Measurement		
		*Number of days	Roots after 2 weeks	
			Number	Length
IBA	0.1	7	4.67	2.13
	0.3	8	3.00	1.40
	0.5	6	9.33	0.73
	1.0	7	10.33	0.43
Mean of		7	6.83	1.17
NAA	0.1	8	3.67	2.00
	0.3	6	4.50	1.25
	0.5	10	5.33	1.67
	1.0	10	3.33	0.57
Mean of		8.8	4.20	1.37
IAA	0.1	8	5.33	1.23
	0.3	9	2.00	0.53
	0.5	9	3.67	1.29
	1.0	9	1.70	0.30
Mean of		8.8	3.18	0.84
L.S.D at 5%		0.87	3.72	0.92

- **Number of days until appearance of visible formed roots per explant**

Acclimatization stage.

6. Effect of transplanting media on survival percentage (%), increase in leaves number per shoots, shoots length (cm), and internodes length(cm) of *Dendranthema grandiflorum* after 4 from transplants.

Data described in Table (6) cleared that, culturing the plantlets on peat moss augmented with perlite and vermiculite (1:1:1v/v) significantly increased the survival percentage as it reached the highest significant value of 100 %.

Also, using the mixture of peat moss + perlite + vermiculite (1:1:1v/v) stimulated the increase of leaves number per shoot, as it reached (5.00 leaves) at the end of adaptation period.

Table 6. Effect of acclimatization media on survival percentage (%), increase in leaves number (leaf), shoots length (cm) and increase in internodes length (cm) after 4 weeks of *Dendranthema grandiflorum transplants.*

Acclimatization media (1;1;1 v/v)		Survival percentage (%)	leaves number per shoot	Increasing of Shoots length (cm)	internodes length (cm)
Peat moss+	Sand	95	3.33	3.93	0.30
	Loamy soil +	93	3.33	3.27	0.33
	Vermiculite Perlite+	100	5.00	6.60	0.53
L.S.D (5%)			2.21	2.20	0.15

Data reported in the same Table revealed that, the mixture of peat moss + perlite + vermiculite was the superior treatment for increasing the shoots and the internodes length comparing with the other mixtures. The transplants grown on a medium containing peat moss + perlite + vermiculite (1:1:1v/v) formed significantly the tallest shoots and internodes (6.60 and 0.53 cm) than the other mixtures, respectively. these

results may be attributed to the interaction effects of the three transplanting media, as presence of vermiculite saved the nutrient elements (containing potassium and magnesium for the last one) in an available form. In addition, presence of peat moss increased the aeration around the plantlets roots.

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تأثير بعض منظمات النمو النباتية علي تكوين الأفرع والجذور للأجزاء النباتية للأراولا المزروعة معمليا.

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أجري هذا البحث بهدف دراسة تأثير التركيزات المختلفة من البنزيل امينو بيورين (BAP) بتركيز (٠.٥ ، ١، ٢ و ٥ ملليجرام/ اللتر) مع نفثالين حامض الخليك (NAA) بتركيز (٠.١ و ٠.٥ ملليجرام/ اللتر) المضاف الي بيئة البداية ، والبنزيل امينو بيورين (BAP) بتركيز (٠.٢ ، ١، ٢ و ٤ ملليجرام/ اللتر) مع نفثالين حامض الخليك (NAA) بتركيز (٠.١ و ٠.٢ ملليجرام/ اللتر) المضاف الي بيئة التضاعف ، وكذلك أنواع مختلفة من الأوكسينات (أندول حمض البيوتريك (IBA) و نفثالين حامض الخليك (NAA) وأندول حمض الخليك بتركيزات (٠.١ ، ٠.٣ ، ٠.٥ و١ ملليجرام/ اللتر) بشكل منفصل المضافه الي بيئة التجذير علي مدي استجابة القطع الساقية لنبات الأراولا إلي محتويات البيئة الغذائية وأقلمة النباتات وكان صنف الأراولا المستخدم في هذا البحث هو صنف فلايروفصلت الأجزاء النباتية المعقمه التي تحتوي علي عقدة واحده بطول (٠.٥-١.٠ سم) وتم زراعتها علي بيئة الزراعة المحتويه علي بيئة مورشيج وسكوج (١٩٦٢) وسكروز٣٠ جم /اللتر و اجار ٧ جم /اللتر و نقلها من بيئة البداية الي بيئة التضاعف ثم الي بيئة التجذير واظهرت النتائج المتحصل عليها في بيئة البداية المضاف اليها البنزيل امينو بيورين (BAP) بتركيز (١.٠ ملليجرام/ اللتر) مع نفثالين حامض الخليك (NAA) بتركيز (٠.٥ ملليجرام/ اللتر) أعلي متوسط لعدد الأفرع كما أن البيئة المضاف اليها البنزيل امينو بيورين (BAP) بتركيز (٢.٠ ملليجرام/ اللتر) مع نفثالين حامض الخليك (NAA) بتركيز (٠.١ ملليجرام/ اللتر) سجلت أعلي طول للأفرع والسلاميات بالأضافة إلي البيئة المضاف اليها البنزيل امينو بيورين (BAP) بتركيز (١.٠ ملليجرام/ اللتر) مع نفثالين حامض الخليك (NAA) بتركيز (٠.١ ملليجرام/ اللتر) اعطت اعلي متوسط لعدد الأوراق. وأيضا أشارت النتائج المتحصل عليها في بيئة التضاعف المضاف اليها البنزيل امينو بيورين (BAP) بتركيز (١.٠ ملليجرام/ اللتر) مع نفثالين حامض الخليك (NAA) بتركيز (٠.٢ ملليجرام/ اللتر) أعلي متوسط لعدد الأفرع والبيئة المضاف اليها البنزيل البنزيل امينو بيورين (BAP) بتركيز (١.٠ ملليجرام/ اللتر) مع نفثالين حامض الخليك (NAA) بتركيز (٠.١ ملليجرام/ اللتر) اعطت أعلي طول للأفرع والسلاميات بالأضافة إلي الافرع النامية في البيئة المضاف اليها البنزيل امينو بيورين (BAP) بتركيز (٠.٢ ملليجرام/ اللتر) مع نفثالين حامض الخليك (NAA) بتركيز (٠.١ ملليجرام/ اللتر) اعطت اعلي متوسط لعدد الأوراق. واخيرا تم نقل النباتات الناتجه من بيئة التضاعف اللي بيئة التجذير المكونة من بيئة مورشيج وسكوج نصف قوة والمزودة بأندول حامض البيوتريك (IBA) بتركيز ١.٠ ملليجرام / اللتر لأنتاج اكبر عدد من الجذور تليها بأندول حامض البيوتريك (IBA) بتركيز ٠.١ملليجرام / اللتر لأنتاج اطول جذور. ثم أقلمة النباتات ونقلها مخلوط الأقلمة (البيت موس : البيرليت : الفيرميكيوليت (١:١:١ بالحجم) للحصول علي اعلي نسبة مئوية للبقاء وعدد الأوراق وطول الأفرع والسلاميات واظهرت تفوقها عن البيئات الاخري.