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EFFECT OF SOME SUPPORTING TYPE, IBA CONCENTRATION AND THEIR INTERACTION ON ROOTING OF *In vitro* REGENERATED MICRO-SHOOTS OF *Polygala myrtifolia* L.

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ABSTRACT

A laboratory experiment was carried out at the Tissue Culture Laboratory, Horticulture Research Institute, Agricultural Research Center, during 2008/2009 to induce rooting on the *in vitro* regenerated micro-shoots of *Polygala myrtifolia* L. on MS medium. These shoots were supported in the culture using four gelling agents (agar at 7.0 g/l, gelrite at 2.0 g/l and agar+gelrite (at 3.5+1.0 g/l, respectively), in addition to a filter paper bridge on a liquid MS medium. The micro-shoots were presoaked in different IBA concentrations (0.0, 0.5, 1.0 and 1.5 ppm) before inoculation on a growth regulator-free 3/4 MS medium.

Results indicate that rooting % and total chlorophyll content increased significantly in agar + gelrite solidified medium, while gelrite significantly increased number of roots/plantlet. IBA at 1.0 ppm significantly increased rooting %, the number of roots/plantlet, root length, total chlorophyll, indoles and total soluble sugars %. The combined effect of the supporting type and IBA concentration exerted a significant effect. Rooting % increased to the significantly highest record when using agar + gelrite in a medium free of IBA.

Therefore, from an economical point of view, it can be recommended in the rooting stage to use agar + gelrite (at 3.5+1.0 g/l MS medium, respectively) without presoaking in IBA solutions to obtain the highest rooting %.

Key words: *agar, filter paper bridge, gelrite , IBA , in vitro, Polygala myrtifolia, rooting.*

1. INTRODUCTION

Myrtle leaf (*Polygala myrtifolia* L., Fam. Polygalaceae), native to South Africa is one of many species of trees and shrubs used in the garden landscape. It is a decorative evergreen shrub; grows to 2 meters in height with crowded, slender branches forming a willowy, oval-shaped bush. The pale purple flowers are pea-shaped, produced in profusion at the tips of the slender branchlets with long flowering period, extending from early spring through the winter when many other plants are not flowering, (Oakman, 1995 and Iapichino, 2004). On the other hand, using standard methods of propagation, *P. myrtifolia* cuttings are slow and difficult to root (Iapichino and Airo, 2002). Consequently, micropropagation might be more effective for the rapid and mass propagation of *P. myrtifolia*.

Many factors affect rooting *in vitro* as gelling agents, auxin type and concentration, media strength, other chemical additions and incubation conditions. Concerning the gelling agents, Ebrahim and Ibrahim (2000) found that agar and gelrite applied (at 5 and 1.5 g/l, respectively) induced rooting of *Maranta leuconeura*. They also stated that adding agar plus gelrite at the rate of 5 + 0.5 g/l, respectively, extremely enhanced rooting. Orlikowska *et al.* (2000) found that diluting MS medium by half when combined with 2 mg/litre gelrite instead of 7 g/litre agar stimulated root growth of *Codiaeum variegatum* cv. Excellent shoots produced *in vitro*. Cao and Earle (2003) showed that the use of a 1:1 mixture of Agar gel and Gelrite in the rooting medium increased the number of healthy roots per rooted plantlet of transgenic broccoli explants. AbdShukor *et al.*

(2008) cleared that MS medium supplemented only with 0.24% gelrite was the most effective treatment producing the longest root length of *Azadirachta excelsa*. In the same manner the positive effect of a liquid medium supported with filter paper was observed by many research workers. Zeng *et al.* (2009) reported that eighty percent of the shoots rooted, and an average of 2.0 roots per shoot was achieved when a basal portion of *Citrus reticulata* Blanco regenerated shoots was dipped into 1000 mg/l IBA solution for 15 min before placement on a filter-paper bridge. Bhattacharya and Bhattacharyya (2010) stated that emergence and elongation of *Jasminum officinale* L, roots from shoot base was facilitated by placing on the notch of a filter paper bridge.

Some authors noticed that the type and concentration of the used auxin. As well as the method by which auxins are used have a conspicuous effect on the obtained result. Radice (2000) reported that root induction of *Codiaeum variegatum* Blume cv Norma (croton) was observed on a basal medium plus 1 mg/l IBA. Romano *et al.* (2002) achieved rooting of *Ceratonia siliqua in vitro* on a growth regulator free medium after basal dipping of shoots in IBA at 4.9 mM (1 ppm) instead of inclusion of auxins in MS medium. In the same regard Iqbal *et al.* (2003) showed that roots of proliferated shoots of 'Hybrid Tea' rose cultivars Rosy Cheek and Whisk Mac were only regenerated in MS medium supplemented with 0.5 mg IBA/liter. Romano and Martins-Loucao (2003) reported that the rooting percentage of *Quercus suber* was improved to 95% by basal immersion of shoots in 0.5 g/litre IBA for 2 minutes. Behera and Thirunavoukkarasu (2006) stated that rooting of *in vitro Desmodium gangeticum* shootlets was better in 1.0 mg/l IBA-supplemented medium than IAA. Burasheed *et al.* (2006) found that the highest rooting percentage was obtained with half-strength MS medium supplemented with IBA at 3.0 mg/litre for date palm cv. Barhee, while for cv. Khalas, 3.0 or 5.0 mg/litre produced the highest rooting percentage and root number/shoot. The longest roots were obtained with IBA at 1.0 mg/litre. Hung *et al.* (2006) stated that root formation of *Wasabia japonica* (Miq.) was greatest with IBA when compared with IAA and NAA. Mehta *et al.* (2007) mentioned that well developed root system of carnation (*Dianthus caryophyllus*) was obtained *in vitro* on a liquid MS medium supplemented with 2.0 mg/l IBA and 0.2 % activated charcoal. Nisha *et al.* (2009) reported that root induction on shoots of *Begonia malabarica in vitro* was achieved on a full strength MS with IAA/IBA at different concentrations (0.0-2.88 mg/l). The regenerated shoots were rooted on MS with 1.2 mg/l IBA. Waseem *et al.* (2009) found that rooting response of chrysanthemum was obtained in a half strength MS medium supplemented with 0.2 mg/l indole butyric acid (IBA).

2. MATERIALS AND METHODS

This experiment was carried out at the Tissue Culture Laboratory, Horticulture Research Institute, Agricultural Research Center, during 2008/2009 season to induce root formation of *Polygala myrtifolia* L. *in vitro* regenerated micro-shoots by using different support type, IBA concentrations applied as pre-soaking and their combinations.

2.1. Explant preparation

Actively growing terminal shoots, 10 cm in length were obtained from 2-year-old *Polygala myrtifolia* plants grown in pots in the experimental nursery of Ornamental Plant Researches and Landscape Department,A.R.C., Giza. The leaves were completely removed, the bare shoots were rinsed under running tap water for about 20 minutes, immersed for 15 minutes in 20% Clorox solution (4-5% sodium hypochlorite) for disinfection and finally rinsed 3 times with sterilized distilled water. Under aseptic conditions, these shoots were cut into one node segments (1.5 cm length) and placed vertically on MS basal medium supplied with 30 g/l sucrose, 7 g/1 agar and 0.4 mg/l BA. Four weeks later, the new *in vitro* produced shoots were transferred to fresh media (with the same previous composition) for multiplication in order to obtain multiple regenerated micro-shoots. After a similar interval, the *in vitro* regenerated micro-shoots (1.5 cm long with intact apices and 3-4 leaves) were moved forward to the rooting stage where they were subjected to different rooting treatments.

2.1.1. Cultural medium and incubation conditions

Murashige and Skoog (1962) nutrient medium (MS) was used at 3/4 strength. This medium contained, in addition to the prescribed salts and vitamins, with 30 g/l sucrose and was solidified with different gelling agents according to the experimental scheme. It was then adjusted to pH 5.8, poured in glass jars (11.5 cm height × 6.5 cm diameter with their polypropylene caps)

and autoclaved at 121°C for 20 minutes under 1.05 kg/cm^2 pressure, left to cool and stored at 25±2ºC for one week before being used. After shoots were inoculated in the medium, the jars were incubated in growth chambers at 25ºC under 16 hrs. photoperiod using Philips cool-white fluorescent tubes at light intensity of 3000 Lux (about 36.3 µmol/m^2/s) till the data were collected about 8 weeks later.

2.2. Experimental treatments

2.2.1. Support type

In order to support shoots in the culture medium, this medium has to be solidified by a gelling agent. However, liquid medium can be used, provided a certain kind of support is available. In this regard, a filter paper can be used to avoid submersion of the explant. In the current study, shoots were inoculated on three different solid media, in addition to a liquid one. The first two media were solidified by either agar at 7.0 g or gelrite at 2.0 g/l MS medium. Gelling was achieved in the third medium by a combination of agar at 3.5 g and gelrite at 1.0 g/l MS medium. In the fourth treatment where a liquid MS medium was used, the explant was supported with filter paper bridge (made of Whatman No. 9).

2.2.2. IBA concentration

In vitro microshoots were soaked before being cultured in the medium in either one of four IBA concentrations (0.0, 0.50, 1.00 and 1.50 ppm) for 2 minutes according to Romano and Martins-Loucao (2003).

At the end of this experiment, the following data were recorded: survival percentage, rooting percentage, number of roots/plantlet, root length (cm), total chlorophyll (mg/100 g F.W.) according to Moran (1982), total indoles (mg/100 g F.W.) as described by A.O.A.C. (1990) and the percentage of total soluble sugars according to Dubois *et al.* (1956). Data were recorded after 8 weeks.

2.2.3. Statistical analysis

The design used was a completely randomized design in a factorial experiment as described by Snedecor and Cochran (1980) at 5% probability level. The supporting agent represents the first factor (four levels), while IBA concentrations represent the second one (four levels). Each one of the 16 treatments consisted of 3 replicates, with 6 jars in every replicate. Data obtained were statistically analyzed using MSTAT Computer Program (1985) and means were compared by Duncan's Multiple Range Test (1955) to verify differences among the means of various treatments.

3. RESULTS

Effect of gelling agents, IBA concentration and their interaction on

3.1. Survival percentage

It can be observed from Table (1) that the supporting type significantly influenced survival percentage of *P. myrtifolia in vitro* regenerated micro-shoots during rooting stage. Gelrite and agar+gelrite significantly induced the highest values in this respect (74.78 and 79.22%, respectively), compared to agar or the filter paper bridge support. It can be also observed that the effect of agar+gelrite was higher than that of gelrite. However, the difference was not significant.

Presoaking the *in vitro* produced *Polygala* shoots in IBA solutions decreased the survival% significantly. This decrement was related significantly and inversely to IBA level. The more the IBA level increased, the less the survival% was. The highest significant value in this regard was obtained when the shoots were not presoaked (87.31%).

The interaction between supporting type and IBA concentrations was found to be significant. Data in Table (1) show that inoculating on agar or agar+gelrite, without presoaking in IBA solution, resulted in the survival of all inoculated shoots. This was followed, without significant difference by presoaking in IBA at 0.5 ppm before inoculating on agar+gelrite (90.28%).

3.2. Rooting percentage

Rooting % increased to the significantly highest record when using agar+gelrite (54.87%) as presented in Table (2). On the other hand, agar presented the next value of rooting percentage, while gelrite and filter paper recorded the lowest values.

The effect of using different IBA concentrations on rooting percentage was significant. It can be noticed that the significantly highest rooting percentage (51.60%) was recorded when shoots were presoaked in IBA at 1.0 ppm. Increasing or decreasing IBA concentration over the former level significantly reduced rooting percentage.

With regard to the interaction between supporting type and IBA concentrations on rooting percentage, the significantly highest record (71.43%) was achieved by using agar+gelrite without IBA as a presoaking treatment, followed without significant

Table (1) : Effect of supporting type and IBA concentration on survival% of *Polygala myrtifolia* **after 8 weeks**

IBA conc. (ppm)	Supporting type				
	Agar	Gelrite	Agar+gelrite	Filter paper	Mean
0.0	100.00 a	79.44 bc	100.00 a	69.81 c-f	87.31 A
0.5	61.51 e-g	76.59 cd	90.28 ab	65.08 d-g	73.36 B
1.0	59.05 f-h	73.81 c-e	71.43 c-f	57.94 f-h	65.56 C
1.5	45.45 h	69.26 c-f	55.19 gh	45.83 h	53.93 D
Mean	66.50 B	74.78 A	79.22 A	59.67 C	

*** Means with the same letter are not significantly different at 0.05 level of significance.**

Table (2): Effect of supporting type and IBA concentration on rooting% of *Polygala myrtifolia* **after 8 weeks**

IBA conc. (ppm)	Supporting type				
	Agar	Gelrite	Agar+gelrite	Filter paper	Mean
0.0	25.92 gh	37.78 e-h	71.43 a	24.44 h	39.89 B
0.5	28.89 gh	24.52 h	50.00 c-f	50.95 b-f	38.59 B
1.0	66.67 ab	41.11 d-g	62.50 a-c	36.11 e-h	51.60 A
1.5	52.22 b-e	55.12 a-d	35.55 f-h	23.33 h	41.56 B
Mean	43.42 B	39.63 BC	54.87 A	33.71 C	42.91

*** Means with the same letter are not significantly different at 0.05 level of significance.**

differences with IBA at 1.0 ppm+agar, IBA at 1.0 ppm+agar+gelrite and finally IBA at 1.5 ppm+gelrite. The lowest rooting% was obtained when the shoots were presoaked in IBA at 1.5 ppm before being supported by a filter paper bridge (23.33%).

3.3. Number of roots/plantlet

Data in Table (3) show that although there was no significant difference between agar, gelrite and agar+gelrite in increasing this trait, these three treatments gave significantly higher values, compared with the result of using filter paper bridge as a support tool. Among the previous mentioned three treatments, gelrite seem to be more effective in increasing this character compared to other treatments.

Presoaking in IBA at 1.0 or 0.5 ppm significantly produced the highest number of roots/plantlet (5.03 and 4.70, respectively), compared to the other treatments. The lowest number of roots/plantlet was found on the untreated plants (3.80).

The interaction between supporting type and IBA concentrations was significant. Agar+IBA (either at 1.0 or 0.5 ppm) significantly increased the number of roots/plantlet to the significantly highest level, with the first combination (5.89) being higher than the second one (5.44). On the other hand, using filter paper bridge either without IBA or with IBA at 1.5 ppm gave the significantly lowest records (3.42 and 3.44, respectively).

3.4. Root length

Results in Table (4) show the effect of supporting type on root length of *P. myrtifolia* plantlets. The significantly longest roots were produced when a filter paper bridge was used to support the shoots on the liquid medium (6.51 cm). Other values produced by other supporting means, *i.e.* gelrite, agar+gelrite and agar came significantly in the second, third and fourth categories (4.93, 4.55 and 3.76 cm), respectively, with significant differences in between. Agar resulted in the shortest roots.

The effect of IBA concentrations was found to be significant. IBA at 1.0 ppm significantly recorded the longest root (5.55 cm). Values of root length at other concentrations came in the second position, with no significant difference in between.

A significant interaction was observed between supporting type and IBA concentrations. The combined treatment of filter paper bridge+ IBA at 1.0 ppm significantly produced the longest roots (7.78 cm) compared with other treatments, followed in the second rank by the filter paper+ IBA at 0.5 (6.67 cm) or with no IBA (6.56 cm). Roots produced on agar with no IBA were significantly the shortest (2.44 cm).

3.5. Total chlorophylls

The effect of different supporting types and IBA concentrations on the total chlorophylls was significant (Table 5). Agar+gelrite proved to be superior in enhancing total chlorophyll content in plantlet leaves as it gave the significantly highest record in this concern (3.51 mg). The

Table (3): Effect of supporting type and IBA concentration on the number of roots/plantlet of *Polygala myrtifolia* after 8 weeks

IBA conc. (ppm)	Supporting type				Mean
	Agar	Gelrite	Agar+gelrite	Filter paper	
0.0	2.55 f	4.35 cd	4.89 bc	3.42 e	3.80 B
0.5	5.44 ab	4.64 bc	4.28 cd	4.42 cd	4.70 A
1.0	5.89 a	4.67 bc	4.83 bc	4.72 bc	5.03 A
1.5	4.22 c-e	4.97 bc	3.61 de	3.44 e	4.06 B
Mean	4.53 A	4.66 A	4.40 A	4.00 B	

*** Means with the same letter are not significantly different at 0.05 level of significance.**

Table (4): Effect of supporting type and IBA concentration on root length (cm) of *Polygala myrtifolia* after 8 weeks

IBA conc. (ppm)	Supporting type				Mean
	Agar	Gelrite	Agar+gelrite	Filter paper	
0.0	2.44 f	5.00 cd	4.67 cd	6.56 b	4.67 B
0.5	3.42 e	4.79 cd	4.39 d	6.67 b	4.82 B
1.0	4.88 cd	5.22 c	4.32 d	7.78 a	5.55 A
1.5	4.29 d	4.71 cd	4.81 cd	5.03 cd	4.71 B
Mean	3.76 D	4.93 B	4.55 C	6.51 A	

*** Means with the same letter are not significantly different at 0.05 level of significance.**

Table (5): Effect of supporting type and IBA concentration on total chlorophyll content (mg/100g F.W.) of *Polygala myrtifolia* after 8 weeks

IBA conc. (ppm)	Supporting type				Mean
	Agar	Gelrite	Agar+gelrite	Filter paper	
0.0	2.78 c	2.42 c-e	2.26 d-f	2.55 cd	2.50 C
0.5	3.33 b	2.04 ef	2.34 c-f	3.33 b	2.76 B
1.0	2.77 c	3.54 b	4.56 a	3.39 b	3.57 A
1.5	1.93 f	4.47 a	4.87 a	3.68 b	3.74 A
Mean	2.70 C	3.12 B	3.51 A	3.24 B	

*** Means with the same letter are not significantly different at 0.05 level of significance.**

significantly lowest one belonged to plantlets grown on agar (2.70 mg). Values of other supporting types; *i.e.,* gelrite and filter paper occupied significantly the second position.

Shoots presoaked in IBA at 1.5 or 1.0 ppm prior to culturing on growth regulators-free MS medium had significantly the highest content of total chlorophylls (3.74 and 3.57 mg, respectively). The same content of shoots grown on a medium supplemented with IBA at 0.5 ppm occupied the second position, while that of shoots deprived of IBA treatment was significantly the lowest.

The interaction between supporting types and IBA concentrations was significant. Agar+gelrite + IBA at 1.5 or 1.0 ppm in addition to gelrite+IBA at 1.5 ppm (4.87, 4.56 and 4.47 mg, respectively) resulted in the significantly highest total chlorophyll content compared to the corresponding records of other combined treatments. The significantly lowest total chlorophyll content was found in plantlets supported with agar+IBA at 1.5 ppm, gelrite with IBA at 0.5 ppm and agar+gelrite with no or 0.5 IBA treatment.

3.6. Indole content in plantlets

The effect of different supporting type on indole content as shown in Table (6) is significant. Agar significantly produced the highest content (4.59 mg), while aga+gelrite scored the significantly lowest record (2.81 mg) in the same regard. The other two treatments resulted in values occupying the second position, significantly.

IBA at 1.0 and 1.5 ppm produced the highest significant indole content, with IBA at 1.5 ppm more effective than IBA at 1.0 ppm (4.22 and 4.00 mg, respectively) but the difference was not significant. The untreated control shoots and those treated with IBA at 0.5 ppm achieved lower results in this concern.

The interaction between supporting type and IBA concentrations was significant. Agar plus IBA at 1.5 ppm significantly produced the highest indole content (7.69 mg). The second category value was recorded by gelrite plus IBA

Table (6): Effect of supporting type and IBA concentration on indole content (mg/100g F.W.) of *Polygala myrtifolia* **after 8 weeks**

IBA conc. (ppm)	Supporting type				Mean
	Agar	Gelrite	Agar+gelrite	Filter paper	
0.0	2.30 gh	3.30 de	2.88 d-f	5.21 c	3.42 B
0.5	3.12 d-f	6.50 b	2.96 d-f	2.04 h	3.66 B
1.0	5.24 c	2.75 e-g	2.69 fg	5.32 c	4.00 A
1.5	7.69 a	3.11 d-f	2.72 fg	3.34 d	4.22 A
Mean	4.59 A	3.92 B	2.81 C	3.98 B	

*** Means with the same letter are not significantly different at 0.05 level of significance.**

Table (7): Effect of supporting type and IBA concentration on total soluble sugars%of *Polygala myrtifolia* **after 8 weeks**

IBA conc. (ppm)	Supporting type				Mean
	Agar	Gelrite	Agar+gelrite	Filter paper	
0.0	2.06 k	2.86 ij	2.90 ij	8.41 c	4.06 d
0.5	2.34 jk	4.38 g	6.17 e	9.20 b	5.52 c
1.0	2.98 i	5.20 f	6.81 d	10.78 a	6.44 a
1.5	3.96 gh	3.45 hi	6.15 e	10.55 a	6.03 b
Mean	2.84 d	3.97 c	5.51 b	9.74 a	

*** Means with the same letter are not significantly different at 0.05 level of significance.**

at 0.5 ppm. The lowest values were those resulted by filter paper bridge+IBA at 0.5 ppm (2.04 mg).

3.7. Total soluble sugars

The total soluble sugars % increased significantly to the highest level by using the filter paper as a supporting tool in the liquid medium (9.74%). On the other hand, shoots grown on agar+gelrite had this parameter in the second position (5.51%). The significantly lowest value was induced by culturing on agar medium (2.84%) (Table ,7).

Presoaking *P. myrtifolia* microshoots in IBA at 1.0 ppm produced the significantly highest percentage of total soluble sugars (6.44%), followed by IBA at 1.5 ppm (6.03%). The significantly lowest percentage was a result of using no IBA at all.

The combined treatments of filter paper bridge + IBA (at either 1.0 or 1.5 ppm) produced the significantly highest values (10.78 and 10.55%, respectively) compared with other treatments. The lowest value was obtained when inoculating on agar with no IBA treatment.

4. DISCUSSION

All the previously mentioned results are in agreement with those obtained by some authors. Ebrahim and Ibrahim (2000) found that adding agar plus gelrite at the rate of 5 + 0.5 g/l, respectively, extremely enhanced rooting. Cao and Earle (2003) on broccoli found that the use of a 1:1 mixture of Agar gel and Gelrite in the rooting medium increased the number of healthy roots/rooted plantlet.

Concerning the effect of IBA concentrations, shoots could be induced to root without auxins or with relatively low concentration, Atta-Alla *et al.* (2003) stated that rooting of proliferated shoots of *Bombax malabaricum* was on MS medium containing different IBA concentrations (0.0-3.0 mg/l), IBA at low concentrations resulted in the highest number of developed roots. Hashem *et al.* (2005) found that, increasing IBA concentration affected rooting % of *Conocarpus erectus* negatively. Hongrat *et al.* (2005) ascertained that the medium without plant growth regulators produced the highest average number of roots of *Cryptocoryne cordata*. However, Banilas and Korkas (2007) reported that relatively low concentrations of indole-3-butyric acid (IBA) promoted both the frequency of shoots forming roots and the number of roots per shoot of grapevine (*Vitis vinifera*) cv. Agiorgitiko genotypes. It may be postulated that the high rooting percentage of micro-shoots cultured in growth regulator-free MS medium may be attributed to the high concentrations of endogenous auxins in these cuttings. If this assumption is true, the application of exogenous auxins may have led to super optimal concentrations in plant tissues, with negative effects on rooting.

In conclusion, from the economical point of view, it is recommended during rooting stage to use agar+gelrite (at 3.5 and 1.0 g/l MS medium,

respectively) without presoaking IBA solutions in order to obtain the highest percentage of rooting.

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تأثير بعض طرق التدعيم وتركيزات إندول حمض البيوتريك والتفاعل بينهما على تجذير الأفرع الدقيقة لنبات البوليجالا الناتجة من زراعة الأنسجة

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ملخص

في تجربة معملية تم إجرائها في معمل زراعة الأنسجة، معهد بحوث البساتين، مركز البحوث الزراعية خلال الموسم ٢٠٠٨/٢٠٠٩ لتشجيع تكوين الجذور على الأفرع الدقيقة الناتجة من زراعة الأنسجة لنبات البوليجالا (*Polygala myrtifolia*, L.) تم استخدام طرق تدعيم مختلفة ، منها أربع مواد لصلابة البيئة و هي الأجار (٧ جم/لتر)، الجيلرايت (٢ جم/لتر)، أجار + جيلرايت (٣,٥ + ١ جم/لتر على التوالى) وكذلك دعامة من ورق الترشيح في بيئة سائلة . نقعت الأفرع قبل زراعتها فى تركيزات مختلفة من إندول حمض البيوتريك (صفر، ٠,٥ ، ١,٠ و ١,٥ جزء في المليون) ثم زرعت على بيئة تحتوى على ثلاثة أرباع تركيز أملاح بيئة موراشيج وسكوج وخالية من منظمات النمو.

أوضحت النتائج أن النسبة المئوية للتجذير و محتوى الكلوروفيل الكلي زادا معنوياً في البيئة المحتوية على أجار + جيلرايت، بينما أدى إستخدام الجيلرايت إلى زيادة معنوية فى عدد الجذور/نبتة.

تسبب النقع المبدئي في إندول حمض البيوتريك بتركيز ١ جزء في المليون فى زيادة معنوية للنسبة المئوية للتجذير ، وعدد الجذور/نبتة، وطول الجذر، ومحتوى الكلوروفيل الكلى ومحتوى الإندولات الكلية ومحتوى السكريات الذائبة الكلية.

وفيما يتعلق بالتفاعل بين طريقة التدعيم وتركيزات إندول حمض البيوتريك ، وجد أن إستعمال الأجار + جيلرايت زاد معنوياً من النسبة المئوية للتجذير فى البيئة الخالية من إندول حمض البيوتريك. وبناء على ذلك ومن وجهةالنظر الإقتصادية فإنه ينصح باستعمال الأجار + الجيلرايت (٣,٥ + ١ جم/لتر على التوالى) بدون نقع الأفرع الدقيقة الناتجة من زراعة الأنسجة فى محلول إندول حمض البيوتريك للحصول على أعلى نسبة تجذير.

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