IN VITRO MICROPROPAGATION OF *VITIS VINIFERA* L. IN KURDISTAN REGION OF IRAQ

Aveen M.N. Ahmed[1], **D**iaa A. Ibrahim[2] and Seyran Palabash Uzun[3]
[1] Biology Dept. , Faculty of science, University of Zakho , Kurdistan Region - Iraq.
[2] Scientific Research Center, Faculty of science, University of Duhok , Kurdistan Region - Iraq.
[3] Department of Bioengineering and science, Kahramanmaraş Sütcü imam University, Turkey.
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Abstract

This investigation was carried out at plant biotechnology laboratories of Scientific Research Center in Faculty of Science, University of Duhok, Kurdistan Region of Iraq, during the period from April 2013 to January 2014. The objectives of the study were to investigate the influence of different plant growth regulators in micropropagation on two local cultivars of grapes *Vitis vinifera* L. The most effective explant disinfestations were obtained by using NaOCl in a ratio of 1:1 water for 10 min which was very effective in reducing contamination in the established cultures resulting in a higher percentage of contaminations free cultures. At initiation stage, the highest initiation rate was achieved by using specific concentrations of BA on basal MS media. At multiplication stage, BA alone and BA combined with GA_3 which was showed different responses in the measured parameters. At rooting formation stage two auxins were tested including IAA and NAA to estimate their ability for root formation. The in vitro propagated of grape plantlets were gradually transferred in successful way from lab. To field conditions, which didn't showed any morphological abnormalities.

Keywords*: *In vitro, Micropropagation, *Vitis vinifera* L.

Introduction

Grapevine "*Vitis vinifera* L." belongs to the family *Vitaceae*. Vitaceae comprise woody climbers, vines, trees, shrubs and succulent's trees (Timmons *et al*., 2007; Chen and Wen, 2007) which are important food sources. Vitaceae family is valuable raw material for the production of wine, medicine and perfumery (Gashkova, 2009). Different varieties produce different types of dried fruit; the dried fruits are the raisins (Kishmish), Currants (Zabib, and the small seedless variety of grape also known as Kishmish). The fruit juice can be concentrated and used as a sweetener. Leaves are cooked. *Vitis vinifera* is not found wild in Kurdistan Region of Iraq although it is very near to the region where it is native to it, i.e. the area between Taurus range and the southern shores of Caspian Sea. But it is extensively cultivated throughout Kurdistan, especially in the mountains where grape yards are usually not irrigated, often on mountainsides of gentle slope, open oak forest, and small moist plains.

Micropropagation of grape through tissue culture provides a valuable tool for a more efficient and rapid multiplication of plant. Plant cell and tissue culture offers means not only for rapid mass multiplication of existing stocks but also for the conservation of important, elite and rare plants. Micropropagation of selected *vitis* genotypes can be carried out, among others, by the culture of intact or fragmented shoot apical meristems, axillary-bud, microcuttings or through adventitious bud formation (Heloir *et al*., 1997). However, most efficient protocols have been reported for muscadine or other than *V. vinifera* grapes (This and Graves, 1992; Qiu *et al*., 2004), while studies with cultivars of *V. vinifera* L. have met with less success (Chee and pool,1983; Zatiko and Molnar, 1985).

The use of apices and axillary buds for the *in vitro* propagation of various species and cultivars of Vitis is documented (Gray and Fischer, 1985) .Working on tendril explants of cultivars of grape, reported somatic embryogenesis and low frequency conversion of embryos to plants. Many authors have found that the use of MS medium supplemented with BA at (1-3 mg l^{-1}). Apical meristem explants were successfully taken from cultures grown on MS medium with 5 µM BA. Cytokinins at different concentrations were tested: 5, 10 and 20 µM BA; 10, 20 and 40 µM kin; 0.5, 1 and 5µM TDZ (Gray and Benton, 1991). It has been shown that MS medium was the most suitable medium for multiplication stage and BA was the best cytokinin to obtain higher proliferation rate. The range of BA that was used in MS medium ranged between 0.5-2 $mg1^{-1}$ showed a better response. Generally , micropropagated plants are difficult to transplant

for two primary reasonds: a heterotrophic mode of nutrition and poor control of water loss (Trigiano and Gray, 1996). This investigation was carried out to test the effects of different plant growth regulators on shoot multiplication to establish a micropropagation protocol of grapes cultivars via (shoot tips and axillary buds segments).

Materials and Methods

Several experiments were carried out during the period from April 2013 to January 2014, in plant biotechnology laboratories of Scientific Research Center, Faculty of Science, Duhok University, Kurdistan Region of Iraq to study the effects of growth regulators on *in vitro* propagation of *Vitis vinifera* L.

All culture glasswares were carefully washed with detergent, and distilled water. Before culturing and using the laminar air flow cabinet, the UV light of the laminar was switched on for 24 hours for sterilizing before culturing. Murashige and Skooge (1962) medium was used as basal medium. Stock solutions, for macronutrients, micronutrients, vitamins and growth regulators at (100 ppm) were prepared and used for the preparation of the media. Also, the media was supplemented with 100 $mg1^{-1}$ *myo*-Inositol, 30 g sucrose and 7 g Agar. The medium was dispensed in equal sizes (25 ml) into culture vessels. Finally, they were autoclaved at 121˚C and 1.04 kg/ m² and then left at room temperature for solidifying to be ready for culture after 24 h (Karim, 2008).

Two kinds of explants (apical shoot tips and lateral buds) of 1.5-2 cm long were taken from *Vitis vinifera* L. field plants, the explants were rinsed under running tap water for 30 mins and drops at dishwashing liquid detergent for 5 min then the explants were transferred to the laminar air flow cabinet to complete the following combinations:

A. 5% NaOCl and distilled water (2:1) + drops of tween-20 for 5 min.

B. 5% NaOCl and distilled water (1:1) + drops of tween-20 for 10 min.

C. 5% NaOCl and distilled water (1:2) + drops of tween-20 for 15 min.

The explants were then rinsed with sterilized distilled water three times, for 5 min, followed by removing the ends of explants that exposed to sterilization. The explants were cultured in jars containing 25 ml of MS medium alone and supplemented with (1 $mg1^{-1}$) BA as an initiation media according to Deniel, (2009).

To determine the suitable combination of plant growth regulators for explants (lateral buds) establishments, the sterilized explants were cultured on MS medium supplemented with BA (0.0 and 1.0 $mg1^{-1}$). Because of the limited numbers of grape vine apical shoot tips, the *in vitro* growing shoot segments with two nodes (1-3 cm) were taken as lateral buds explants. Measurements were recorded after 4 weeks including shoot number / explants and mean length of shoot. After 4 weeks of culture, the shoots were cut into segments of shoot tips and two nodes (≥ 1 cm long) and were cultured on multiplication MS medium. Multiplication stage was divided into two experiments, at the first experiment the explants were cultured on MS medium supplemented with BA alone at different concentrations (0.0, 0.5, 1.0, 1.5 and 2.0 mg l^{-1}) and same BA protocols combined with GA_3 (0.25 mg l^{-1}). For the second experiment to determine the effect of these combinations on the studied parameters.

After 4 weeks of incubation in multiplication medium, the plantlet microshoots were transferred to rooting media. To determine the most suitable auxins concentrations and salt strength of MS media for rooting, microshoots were cultured on:

A. MS medium supplemented with different concentrations of NAA (0.0, 0.5, and 1.0 mgl^{-1}).

B. MS medium supplemented with different concentrations of IBA (0.0, 0.5, and 1.0 mgl^{-1}).

After 4 weeks, data were recorded including rooting percentage, number of root/explant and mean length of roots. For all the treatments, the cultures were incubated in the growth room at 24±1˚C under 16 h photoperiod by white fluorescent tubes under light intensity of 1000 lux. Three replicates were cultured in jars containing 25 ml of MS medium.

The rooted plantlets were taken from rooting media and washed thoroughly with water to remove adhering media, immersed in a beaker containing 1 gml^{-1} Benlet fungicide for 10 mins. and washed with distilled water then the rooted plantlet were transferred to pots containing autoclaved peatmoss, loam and Styrofoam (1:1:0.5) (v:v:v). The pots were placed in sterilized box and covered by polyethylene cover during the first week. The post were enclosed in polyethylene bags, which were closed and placed in a shaded area of temperature – controlled greenhouse set at 23-25c. The plants

were irrigated with a nutrient solution containing 1/4 strength of MS salts. After 8 to 10 days, the bags were opened and after another 8 to 10 days, the bags were removed and plants were grown under regular greenhouse condition (Toma, 2009).

The experiments were arranged according to Complete Randomized Design (C.R.D) using three replicates for each treatment. Data scored in percentage were subjected to arcsine transformation before statistical analysis and then converted back to percentages for presentation. Data were analyzed and means were compared with each other using Duncan's multiple range test to evaluate the significant differences between the means.

Results and Discussion

The results showed that the combination (B) which have been used in surface sterilization of both types of explants gave the highest percentage of healthy - uncontaminated- explants which was achieved 70 % in Be-dandk cultivar ,while was reached 66 % in apical shoot tips of Des-alaneez cultivar. On the other hand, healthy lateral bud explants showed a higher initiation response (95 %) in Be-dandk cultivar, followed by (90 %) in Des-alaneez lateral buds cultivar. Therefore, the second treatment was selected for disinfesting the explants for later experiments.

As it is known that the minimum concentration and duration of sterling treatment is more preferable to insure not damaging the treated explant (Razdan, 2003). Sodium hypochlorite proved to be the best sterilant ever being effective not only as decontaminant but also easy to remove from explants in a minimal damage to the explant tissue. And the sterilization was efficient in eliminating various pathogens such as bacteria and fungi, as well as prevention of browning of explant in grapevine tissue culture (Dalal *et al.,* 1991). The higher initiation response in lateral buds it might be due to the higher content of endogenous hormones. These results are seems to be similar to those obtained by Razdan (2003) and Amin (2007).

At multiplication stage, microshoots produced at initiation stage were used as explants sources for shoot multiplication stage. The ability of different concentrations of BA (0.0, 0.5, 1.0, 1.5, and 2.0 mg l^{-1}) alone and same levels of BA supplemented with (0.25 mg l^{-1}) GA_3 were used to induce *Vitis vinifera* L. shoot multiplication.

Table (1): Effect of different BA concentrations and BA+ GA_3 combination on highest shoot length (cm) of Grapevine plants after four weeks in culture .

Cultivars	Hormones	Hormone concentration mgl^{-1}					Effect of cultivar × hormone	Effect of cultivars
		0	**0.5**	**1.0**	**1.5**	**2.0**		
Be- dandek	**BA**	1.43c	2.27ab	2.20ab	2.70a	1.63bc	2.05a	1.60a
	BA + GA_3	0.70c	1.18 ac	1.48a	1.42ab	0.97bc	1.15c	
Des –alaneez	**BA**	1.15b	1.57ab	1.93a	2.07a	1.27b	1.60b	1.46a
	BA + GA_3	1.37a	1.67a	1.23a	1.15a	1.20a	1.32c	
Effect of cultivar × concentration	**Bedandek**	1.07e	1.73bc	1.84ba	2.06a	1.30ed		
	Des alaneez	1.26ed	1.62bc	1.58dc	1.61bc	1.23e	**Hormone Effects**	
Effect of hormone × Conc.	**BA**	1.29c	1.92b	2.07b	2.38a	1.45c	1.82a	
	BA+GA_3	1.03c	1.43c	1.36c	1.28c	1.08c	1.24b	
Concentration effect		1.16b	1.67a	1.71a	1.83a	1.27b		

Different letters represent significant differences according to Duncan's multiple range tests at 5% levels.

Table (1) shows the effects of BA levels and BA enriched with (0.25 mg l^{-1}) GA_3 after four weeks in culture on highest shoot length of grapevine plant.

The effect of the hormones clarify that the media supplemented with different concentration of BA increased significantly and reaches a highest length (1.82 cm) when compared with the BA + GA_3 (1.24 cm).

Table (2) Generally, the data clarify that there is no significant differences between the two cultivars used in this investigation, whereas the effect of cytokinins induce significant increase in this parameter by using the combination of BA with GA_3 (1.09 cm) when compared with the medium supplemented with BA alone (1.01 cm). In the same table, the result elucidate that the shoot length average reaches (1.17 cm) in Be-dandek cultivar because of the interaction between cultivars and plant growth regulators, which was reduced significantly (0.88 cm) by adding BA + GA_3.

Contrastive response has been shown in Des-alaneez cultivar whereas the average shoot length increased significantly in the medium enriched with BA + GA_3 (1.31 cm) while achieve (0.85 cm) in same cultivar cultured in different levels of BA alone.

Table (2): Effects of different BA concentrations and BA+GA_3 combination on shoot length average (cm) of Grapevine plants after four weeks in culture.

Cultivars	Hormones	Hormone concentration mgl^{-1}					Effect of cultivar × hormone	Effect of cultivars
		0	**0.5**	**1.0**	**1.5**	**2.0**		
Be- dandek	**BA**	1.10bc	1.40ab	1.07bc	1.60a	0.70c	1.17a	1.02a
	BA + GA_3	0.41b	0.88ab	1.02ab	1.13a	0.93ab	0.88b	
Des –alaneez	**BA**	0.63bc	0.97ab	0.97ab	1.13a	0.57c	0.85b	1.08a
	BA + GA_3	1.13a	1.30a	1.50a	1.28a	1.35a	1.31a	
Effect of cultivar × concentration	**Bedandek**	0.76c	1.14bac	1.04bac	1.37a	0.82bc		
	Des alaneez	0.88bc	1.13bac	1.23ba	1.21ba	0.96bac	**Hormone Effects**	
Effect of hormone × Conc.	**BA**	0.87bdc	1.18bac	1.02bdc	1.37a	0.63d	1.01b	
	BA+GA_3	0.77dc	1.09bac	1.26ba	1.21ba	1.14bac	1.09a	
Concentration effect		0.82c	1.14ba	1.14ba	1.29a	0.89bc		

Different letters represent significant differences according to Duncan's multiple range tests at 5% levels.

Table (3) declares that the addition of BA levels in general to MS multiplication media was valuable in case of number of shoots per explants after four weeks in culture. Since the highest number of shoots per explants (2.9) in the medium enriched with different levels of BA which decreased significantly (1.78 shoot / explant) in the same concentration of BA supplemented with 0.25 mgl^{-1} GA_3 (Figure 1). On the same manner, both cultivars (Be-dandek and Des alaneez) recorded the highest significant shoot numbers per explants (2.99 , 2.81) respectively when cultured in the media supplemented with BA whereas, achieved (1.8 , 1.77) in the medium containing different concentrations of BA + GA_3. Although that, the data didn't shown any significant differences between the two cultivars.

Tableb(3): Effects of different BA concentrations and BA+GA_3 combination on number of shoots/ explant of grapevine plants after four weeks in culture .

Cultivars	Hormones	Hormone concentration mgl^{-1}					Effect of cultivar × hormone	Effect of cultivars
		0	**0.5**	**1.0**	**1.5**	**2.0**		
Be- dandek	**BA**	1.50b	3.03cb	3.17ba	3.20ba	4.07a	2.99a	2.40a
	BA + GA_3	1.83a	1.67a	2.00a	2.00a	1.50a	1.80b	
Des –alaneez	**BA**	1.10c	2.60bcd	2.73bcd	3.50ab	4.10a	2.81a	2.29a
	BA + GA_3	1.53a	1.83a	1.90a	2.00a	1.56a	1.77b	
Effect of cultivar × concentration	**Bedandek**	1.67bc	2.35ba	2.58a	2.60a	2.78a		
	Des alaneez	1.32c	2.22ba	2.32ba	2.75a	2.83a	**Hormone Effects**	
Effect of hormone × Conc.	**BA**	1.30c	2.82b	2.95b	3.35b	4.08a	2.90a	
	BA+GA_3	1.68c	1.75c	1.95c	2.00c	1.53c	1.78b	
Concentration effect		1.49c	2.28b	2.45ba	2.68ba	2.81a		

Different letters represent significant differences according to Duncan's multiple range tests at 5% levels.

Table (4). It is clear that using BA plant growth regulator in multiplication medium cause highly significant increase in number of leaves / explant (8.19) when compared with BA+GA_3 combination which reduce the number of leaves (4.35) (Figure 1). Although there was no significant differences between the two investigated cultivars in this parameter; The results indicate that the interaction between the cultivars and hormone concentration increase the number of leaves per explant (8.21 and 8.17) in Be-dandek and Des alaneez, respectively in the media containing BA only which was decreased significantly in the same interaction by adding GA_3 (4.82 and 3.89mgl^{-1}) for both cultivars.

Table (4): Effects of different BA concentrations and BA+GA_3 combination on number of leaves / explant of Grapevine plants after four weeks in culture.

Cultivars	Hormones	Hormone concentration mgl^{-1}					Effect of cultivar × hormone	Effect of cultivars
		0	**0.5**	**1.0**	**1.5**	**2.0**		
Be- dandek	**BA**	4.17cb	11.53a	9.00ab	8.97ab	7.40c	8.21a	6.52a
	BA + GA_3	3.33a	4.77a	4.83a	6.00a	5.17ab	4.82b	
Des –alaneez	**BA**	2.47c	10.17a	10.33a	11.60a	6.30bc	8.17a	6.03a
	BA + GA_3	3.43b	3.50b	3.77b	5.43b	3.30b	3.89b	
Effect of cultivar × concentration	**Bedandek**	3.75c	8.15a	6.92ba	7.48a	6.28ba		
	Des alaneez	2.95c	6.83ba	7.05a	8.52a	4.80bc	**Hormone Effects**	
Effect of hormone × Conc.	**BA**	3.32c	10.85a	9.67a	10.28a	6.85b	8.19a	
	BA+GA_3	3.38c	4.13c	4.30c	5.72cb	4.23c	4.35b	
Concentration effect		3.35c	7.49a	6.98ba	8.00a	5.54b		

Different letters represent significant differences according to Duncan's multiple range tests at 5% levels.

Shoot multiplication traits were influenced by addition of BA more than mixing it with GA_3 and this could due to the number of double bonds on its molecule structure (Mohammed, 1985) which is provided with three bonds in its side chain and consequently increase the activity. In addition to presence of benzene ring on BA structure improve its efficiency and become the most effective cytokinin (Wasfy, 1995). Positive role have been seen on multiplication stage (shoot numbers, length and leaves numbers), may be due to two causes:

First, is that the cytokinin increase the enzymes and proteins and RNA synthesis in the cells which promote bud growth (Al-Rifae and Al- Shobaki, 2002). The second, releasing lateral buds from the dominance of terminal buds without the need of removing the apical bud by promoting formation of xylem tissues of buds which facilitate the transporting of water and nutrients. (Mohmmed and Younis, 1991).

One of the major physiological effects of the auxins is the stimulating of adventitious roots formation in both *in vitro* and *in vivo* cutting (Hartmann *et al.,* 2002). At rooting stage, *in vitro* shoots derived from multiplication stage were used and two kind of auxins (IBA and NAA) as followed: MS medium supplemented with different concentration of NAA (0.0, 0.5, and 1.0 mg^{-1}). MS medium supplemented with different concentration of IBA (0.0, 0.5, and 1.0 mg^{-1}). Roots were initiated at rooting formation stage, following 4 weeks of incubation on rooting media of above specific concentrations, thereafter the roots were adventitiously initiated at cut margins of the shoots.

The percentage of root formation was significantly affected by the different treatments tested on *Vitis vinifera* L.. Table (5). It is clear that increase the concentration of investigated auxins from 0.5 to 1.0 mgl^{-1} caused significant increase (82.0% and 86.33%) in rooting percentage of grape plants compared with free media of auxins.

Concerning the differences in rooting abilities of grape, regarding the combination between types of hormones and concentration of auxins, 0.5 and 1.0 $mg1^{-1}$ NAA was nearly similar in both concentration which produced the highest significant rooting percentage (87.83% and 87.33%), respectively when compared with control treatment (68.61%).Whereas 1.0 $mg1^{-1}$ concentration of IBA produced the highest rooting percentage (85.33%) and decreased significantly in control treatment (58.33%). On the other hand, no significant difference was seen between cultivars.

Table (5): Effects of different Auxin (NAA and IBA) concentrations on rooting percentage (%) of grapevine plants after four weeks in culture.

Cultivars	Hormones	Auxin concentration mgl^{-1}			Effect of cultivar × hormone	Effect of cultivars
		0	0.5	1.0		
Be- dandek	**NAA**	68.89bc	95.67a	88.00bc	84.18a	79.20a
	IBA	53.33c	82.00bc	87.33bc	74.22a	
Des –alaneez	**NAA**	68.33bc	80.00bc	86.67bc	78.33a	75.33a
	IBA	63.33bc	70.33a-c	83.33bc	72.33a	
Effect of cultivar × concentration	**Bedandek**	61.11b	88.83a	87.67a		
	Des alaneez	65.83b	75.17ab	85.00a	**Hormone Effects**	
Effect of hormone × Conc.	**NAA**	68.61bc	87.83a	87.33a	81.26a	
	IBA	58.33c	76.17ab	85.33a	73.28a	
Concentration effect		63.47b	82.00a	86.33a		

Different letters represent significant differences according to Duncan's multiple range tests at 5% levels.

Data in Table (6) clarify that there is a significant difference in the number of roots formed on explants of *Vitis vinifera* L. as a result of tested treatments. The highest number of roots/explant (14.44) was recorded in Bae-dandk cultivar compared with Des-alaneez (7.039 roots/explant). Regarding the effects of the auxins on this parameter it is obvious that the effect of NAA was more valuable of IBA which increase the roots significantly number to (13.33 roots / explant) compared with IBA (8.15 root/explant)(Fig 1).

Increasing the concentration of auxin from 0.5 to 1.0 mgl^{-1} increase the roots number (14.13 root/explant) significantly according to control treatment (4.33 roots / explants).

Table (6): Effects of different Auxin (NAA and IBA) concentrations on root numbers/ explants of grapevine plants after four weeks in culture.

Cultivars	Hormones	Auxin concentration mgl^{-1}			Effect of cultivar × hormone	Effect of cultivars
		0	0.5	1.0		
Be- dandek	**NAA**	2.83f	29.67a	26.17a	19.56a	14.44 a
	IBA	2.83f	11.50bc	13.67b	9.33b	
Des –alaneez	**NAA**	4.00ef	8.50cd	8.83cd	7.11b	7.039 b
	IBA	7.67c-e	5.40d-f	7.83c-e	6.97b	
Effect of cultivar × concentration	**Bedandek**	2.83c	20.58a	19.92a		
	Des alaneez	5.83b	6.95b	8.33b	**Hormone Effects**	
Effect of hormone × Conc.	**NAA**	3.42c	19.08a	17.50a	13.33a	
	IBA	5.25c	8.45b	10.75b	8.15b	
Concentration effect		4.33b	13.77a	14.13a		

Different letters represent significant differences according to Duncan's multiple range tests at 5% levels.

Valuable inhibition in the number of roots formation was shown in Des alaneez cultivar. It is clear that both auxins (NAA, IBA) weren't too much effective to increase the roots numbers although of simple increase when compared with control treatments. Combined cultivars with type of auxin significantly increased the number of roots (19.50 roots/explants) which were formed on shootlets of Bae-dandek cultivar when cultured on MS supplemented with NAA compared with Des-alaneez when cultured on MS supplemented with NAA and IBA (7.11, 6.97 roots/explants) respectively.

Root length average was affected significantly as a result of adding investigated auxins to the medium. Table (7) clarify that there was significant increase in root length in Bae dandek cultivar (4.6 cm) compared with Des alaneez (2.27 cm). Hormone effect declares that the IBA induce more significant increase in root length average (5.17 cm) than NAA (1.69 cm).

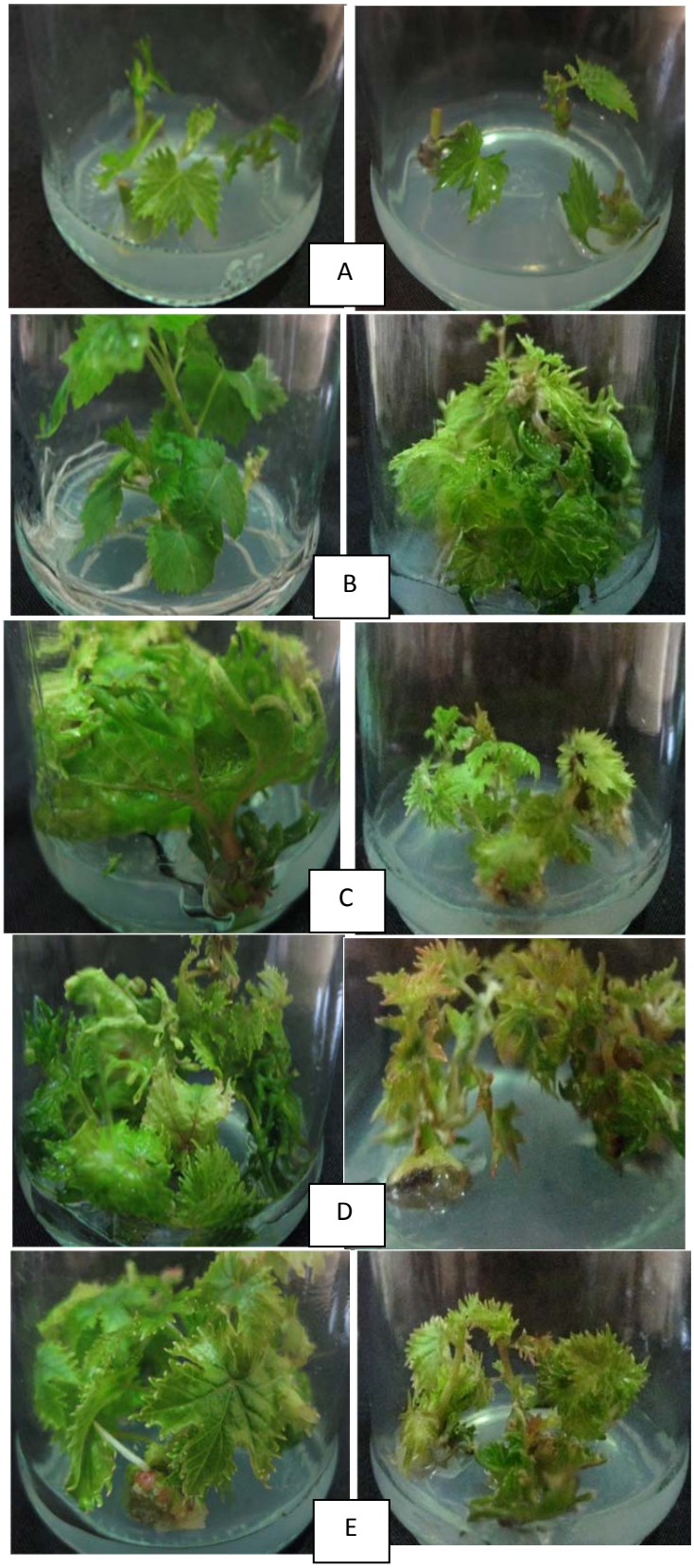
Table (7): Effects of different Auxin (NAA and IBA) concentrations on root lengths average (cm) / explants of Grapevine plants after four weeks in culture.

Cultivars	Hormones	Auxin concentration mgl^{-1}			Effect of cultivar × hormone	Effect of cultivars
		0	**0.5**	**1.0**		
Be- dandek	**NAA**	3.50bc	2.27b-e	2.00b-e	2.59b	4.60a
	IBA	4.00b	7.90a	7.92a	6.61a	
Des –alaneez	**NAA**	0.93c-e	0.67e	0.80de	0.80c	2.27b
	IBA	3.33b-d	4.37b	3.50bc	3.73b	
Effect of cultivar × concentration	**Bedandek**	3.75ab	5.08a	4.96a		
	Des alaneez	2.13b	2.52b	2.15b	**Hormone Effects**	
Effect of hormone × Conc.	**NAA**	2.22bc	1.47c	1.40c	1.69b	
	IBA	3.67b	6.13a	5.71a	5.17a	
Concentration effect		2.94a	3.80a	3.55a		

Different letters represent significant differences according to Duncan's multiple range tests at 5% levels.

Finally the result of root formation stage, indicate that the auxins have an important role in promotins adventitious root establishment on shoot end cuttings (Abdul, 1978; Saleh, 1991).

Adventitious root formation is a complex process that is affected by multiple endogenous factors including phytohormones and environmental factors. Both auxins NAA and IBA varied in their effects on grapevine rooting whereas NAA was more effective in the case of rooting percentage and root number per explants while IBA was preferable in its effect in the stimulation of root length, these results was similar to finding of those of Danial *et al*., (2009). And confirm that the presence of these auxins had positive influence on rhizogenesis under *in vitro* conditions.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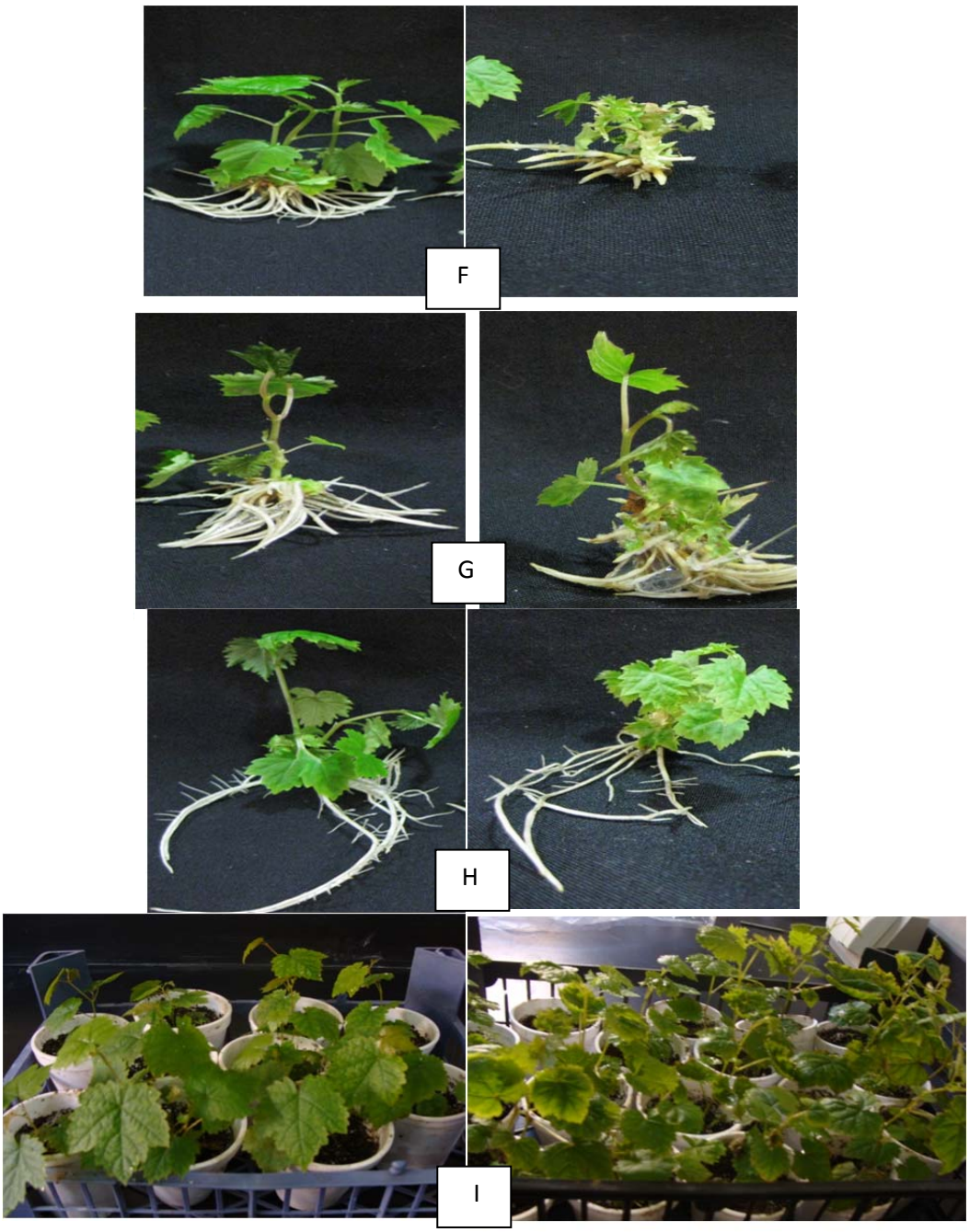

Figure (1):
A. Initiation shoots from lateral buds on MS medium enriched with 1.0 mgl^{-1} of BA after 4 weeks in culture. (Bae dandek , Des alaneez Cultivars respectively in all picture)
B. Multiplication stage: Effects of different BA concentrations and BA+ GA_3 combination on highest Shoot Length (cm) of grapevine plant
C. Multiplication stage: Effects of different BA concentrations and BA+ GA_3 combination on Shoot Length Average (cm).
D. Multiplication stage: Effects of different BA concentrations and BA+ GA_3 combination on number of Shoots / explant.
E. Multiplication stage: Effects of different BA concentrations and BA+ GA_3 combination on number of leaves / explant.
F. Rooting stage. Effect of different IBA and NAA on rooting percentage (%) of Grapevine plants.
G. Rooting stage: Effects of different IBA and NAA on number of roots of Grapevine plants.
H. Rooting stage: Effect of different IBA and NAA on roots length average of Grapevine plants.
I.Acclimatization stag

CONCLUSIONS

It can be concluded that treating the explants with Sodium hypochlorite + distilled water (1:1) was more effective in reducing explants contamination. At initiation stage, BA showed better results for establishing aseptic *Vitis vinifera* culture from lateral buds while in initiation stage, lateral bud explants were more effective in producing shoots than apical bud explants. Hence, shoot multiplication traits were influenced by addition of BA more than mixing it with GA3

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الاكثار الدقيق للعنب المحلي في اقليم كردستان العراق

الخلاصة:

اجريت هذه الدراسة في مختبر الزراعة النسيجية التابعة لمركز الابحاث العلمية /فاكوليتي العلوم /جامعة دهوك /اقليم كردستان العراق، خلال الفترة من نيسان ٢٠١٣ الى كانون الثاني ٢٠١٤. وكان الهدف من هذه الدراسة هو دراسة التأثيرمنظمات النمو على الاكثار الدقيق للأصناف المحلية من العنب المزروعة في اقليم كردستان العراق ، تم الحصول على افضل التعقيم السطحي للبراعم باستخدام محلول القاصر التجاري بتركيز ٥٪ هايبوكلورات الصوديوم لمدة عشرة دقائق بنسبة ١:١ المياه وكانت فعالة جدا في الحد من التلوث في اوساط النمو التي أنشأت. واظهرت النتائج في مرحلة البدء المعدل العالي ويتحقق باضافة تركيزات محددة من(BA)الى وسط القاعدي. وفي مرحلة التضاعف استخدمنا نوعين من سايتوكاينين بما في ذلك(BA) وحده، (BA+GA) ومع الذي اظهرت استجابات مختلفة في المعلمات قياس. وفي مرحلة تكوين الجذور تم اختيار اثنين من الاوكسينات بما في ذلك(NAA,IBA) و لتقدير قدرتهم على تشكيل الجذور. تم نقل الشتلات تدريجيا من مختبر الزرع الى الحقل بطريقة ناجحة ولم يظهر أي تشوهات شكلية في الحقل.

زیدەبونا وردبینی لسەر تریّ یی خومالی ل هەریما کوردستانا عیراقی

کورتی:

ئەڤ ڤەکولینە هاتیە کرن ل لابورا چاندنا شانا یاگریدایێ ب سەنتەری ڤکولینیت زانستی فاکولتیا زانست/ زانکویا دهوك/ هەریما کوردستانا عیّراقی. ل ماوەی هەیڤا نیسانی ٢٠١٣ تا کانوونا دووێ ٢٠١٤.

ئارمانج ژڤی ڤەکولیّنی ئەوە کو کارتیکرنەکا (منڤمات النمو) گەلەك دەقیقە لسەر جوریّت تری ییّن خوّمالی ل هەریما کوردستانا عیّراقی.باشترین ئەنجام هاتن وەرگرتن (التعقیم) سەرڤە بو پشکوژین هاتین بەکارئینان ب بەکارئینانا ٥٪ هایپوکلورات الصودیوم بو ماوەی ١٠ خولەکا بەریژا ١:١ ئاڤ وکارتیکرنا گەلەك دژوار بوو بوّ ژناڤبرنا پیس بوونی لە نلڤ (اوسات النمو). ودیار بوو ئەنجامێقوناغا دەس پیّکا گەشەکرنا پشکوژا یا بلندە بوو ب زیادەکرنا BA بوّ میدیا تفت. ل قوناغا زیادەبوونێدا مە بەکارئینان دوو جوریّت سایتوکاینین لەجارەکیّ BA بتنیّ وجارا دووی BA لەگەل GA3 هاتین بەکارئینان وئەنجامیّن جیاواز هلتنە دیارکرن. ل قوناغا چیبوونا رەگا مە هەلبژارتن دوو جور ژ ئوکسیناتا (NAA,IBA) دا شیانیت وان دیاربن بو جیکرنا رەگا. ودوومایکیّ شتل هاتن ڤەگوهاستن ژ لابورا جاندنی بوّ دەرڤە بریکەکا سهرکەفتی ونەهاتە دیارکرن جیّ (تشوهات شکلی) ل جهیّ هاتین جاندن.