



Studies on the Behavior of Proliferated Shoots and Roots of Two Fig Cultivars *in Vitro*

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Abstract: Shoots regenerated from shoot tip of Black Mission and Conadria fig cultivars plantlets from the establishment stage were cultured on Murashige and Skoog medium. Different cytokinin types (6-benzylaminopurine (BAP), 2ip (6-(γ,γ -Dimethylallylamino)purine) and kinetin) and cytokinin like compound (TDZ) were tested, in addition, Murashige and Skoog (MS) and woody plant (WP) medium were studied on proliferation and rooting stage. Data indicated that culturing of shoot regenerated from shoot tips on woody plant medium supplemented with 0.5 mg/L Kinetin is recommended during the proliferation stage. However, Black Mission cultivar surpassed Conadria cultivar in increasing shoot length and number of roots parameters. Also, number of roots (12.06) and root length (4.45) of fig shoots were better on free woody plant medium compared with those medium contained IBA and free Murashige and Skoog medium.

Key words: *Ficus carica*, Black Mission, Conadria, Shoot multiplication, micropropagation, Cytokinin type – Cytokinin like compound, TDZ and rooting stage.

Introduction

The common fig (*Ficus carica* L.) is one of the first plants ever to be cultivated by human; it is a woody plant with highly varied forms, very tall, small trees, bushes, shrubs and large tree¹. Development of an efficient plantlets regeneration in fig is essential for successful *in vitro* proliferation, rooting and transformation, Fig is recalcitrant in its production of shoot regeneration² and factors affecting shoot multiplication have not been optimized³.

The most important technique of tissue culture reported by various researches using cultures of shoot tips and axillary buds to regenerate multiple shoots^{4,5}. Several authors described merits of tissue culture technique to produce free virus plants, mass production in short duration, valuable tools for breeding programs, good tool to overcome traditional propagation methods and research studies⁶⁻¹².

Meanwhile, *in vitro* proliferation and rooting of *Ficus carica* L. are affected by many factors¹³⁻¹⁶. These factors included culture medium type, medium strength and concentration of growth regulators. The composition of the medium is a determining factor for growth parameters. The cell of most plant species can be grown on completely defined media types and strength, the wide used of Murashige and Skoog medium or its modification. The kinetin showed a less stimulatory effect of multiple shoots when compared to BAP. This can be explained on the basis of two double bonds in 6-benzylaminopurine structure as compared with a single bond in kinetin molecule¹⁷. The highest values of rooting were observed with MS medium supplemented with 1 mg/L IBA in all genotypes i.e. Conadria 73.3%, Deamna 52.2%, Brown turkey 26.7% and Poona 24.4%¹⁸.

Materials and Methods

This study was carried out at the tissue culture laboratory of Pomology Dept., National Research Centre, during the Period from 2013 to 2015.

Shoot tip of Black Mission and Conadria fig cultivars plantlets from the establishment stage were cultured individually on ¹⁹ as a basal medium supplemented with 0.5 mg/L 6-benzylaminopurine (BAP), 30g/L sucrose and 6 g/L Difco-Bacto Agra during the proliferation stage. The pH of the medium was adjusted 5.7 and autoclaved at 121oC and 15 Ib/inc for 15 minutes. The culture explants were incubated under 16 hours of artificial light (Fluorescent light at 30 μ m/sec) and 8 hours of darkness at average temperature at 23+2°C ²⁰. Thus the following experiments were carried out.

1- Effect of cytokinin type, medium type and cytokinin like compound

Kinetin, 6-benzylaminopurine, 2ip (6-(γ,γ -Dimethylallylamino)purine)andThidiazuron (TDZ) were applied to both cultivars¹⁹ and ²¹ medium at 0.5 mg/L level either alone or in combinations to find out the most effective cytokinin to enhance the highest shoot proliferation.

2- Effect of medium type or rooting:

Indole-3-butyric acid (IBA) at 0.5 mg/L was added in¹⁹ and ²¹ medium to find out the best medium which gave the greatest rooting. At the end of experiments, average shoot number, leaf number, shoot length (cm), root length (cm) and number of roots were recorded after 6 weeks of culturing.

3- Statistical Design:

Treatment were arranged in complete randomized design, each treatment were replicated 5 times for each treatment according to²². The obtained data was statistical analyses and the means were differentiated according to Duncan multiple range test at 5% level as described by²³.

Results and Discussions

1. 1. Proliferation stage:

It appearedfrom Table (1-a) that BlackMission cultivar showed highly significant increase in shoot length as compared with Conadria cultivar. On the other hand, number of shoots as well as number of leaves didn't show any statistical differences when two Fig cultivars were used.

Meanwhile, Table (1-b) indicated that using woody plant medium plus 0.5 mg/L kinetin gave the highest significant increase of shoot length and number of leaves as compared with used treatments. However, using Murashige and Skoog ¹⁹ media + 3mg/L BAP + 1 mg/L GA3 as well as WP + 0.5 mg/L Kinetin gave the highest significant increase of number of shoots in relation to MS + 2 mg/L TDZ + 4 mg/L 2ip.

Table (1): Effect of different concentration of cytokinin type and cytokinin like compound on proliferation and growth parameters of two Fig cultivars.

Table (1-a): Effect of cultivar:-

Growth parameter Cytokinin type	Number of shoots (No.)	Shoot length (cm)	Number of leaves (No.)
BlackMission	32.44	2.19a	2.94
Conadria	40.5	1.51b	2.51
L.S.D	NS	0.61	NS

Means followed by the same letter(s) with each parameter are not statistically different 5% level.

Table (1-b): Effect of cytokinin type:-

Growth parameter Cytokinin type	Number of shoots (No.)	Shoot length (cm)	Number of leaves (No.)
MS+2mg/L TDZ + 4 mg/L 2ip	16.58b	1.03b	1.52b
WP + 0.5 mg Kinetin	44.75a	3.04a	3.90a
MS+3mg/L BAP + 0.1mg/L GA3	48.08a	1.48b	2.76ab
L.S.D	12.37	0.74	1.38

Means followed by the same letter(s) with each parameter are not statistically different 5% level.

Dealing with the interaction between Fig cultivars and cytokinin type, Table (1-C) verifies that number of shoots recorded a significant increase when Conadria Fig explants cultured on MS + 3mg/L BAP + 0.1mg/L GA3. On the other hand, statistical differences were lacked among different interactions under study when shoot lengths and number of leaves parameters were discussed. Generally, the aforementioned results indicated that BlackMission cultivar surpassed Conadria cultivar in increasing shoot length parameter. In addition, it could be recommended that Lloyd, G. and B.H. McCown²¹ medium supplemented with 0.5 mg/L Kinetin was the best than other medium types with multiplication; this conclusion within the same Lin with that found by^{2,24}. They reported that the highest proliferation of shoot tip explants of different Fig cultivars using Kinetin. On the other hand,^{25,18} found that the best shoot multiplication of fig cultivars were observed on medium contained BAP, GA3 and IBA.

Table (1-c): Effect the interaction between fig cultivar and cytokinin type

Growth parameter Cultivar	Number of shoots (No.)		Shoot length (cm)		Number of leaves (No.)	
	BlackMission	Conadria	BlackMission	Conadria	BlackMission	Conadria
MS+2mg/L TDZ + 4 mg/L 2ip	15.66	17.50	1.14	0.91	1.92	1.11
WP + 0.5 mg kinin	55.00	34.50	3.52	2.56	4.11	3.96
MS+3mg/L BAP + 0.1mg/L GA3	26.67	69.50	1.92	1.05	2.79	2.74
L.S.D	33.78		NS		NS	

Means followed by the same letter(s) with each parameter are not statistically different 5% level.

1. 2- Rooting stage:-

Data in table (2-A) reflected that Black MissionFig explants showed significant increase in shoot length and number of roots than Conadria Fig. On the other hand, statistical difference were nil between two Fig cultivars root length and number of leaves parameters were concerned.

Moreover, Table (2-b) explains that MS or WP medium free was significantly superior to other medium type in increasing number of roots and root length. Also, shoot length was significantly maximized as WP media free as well as up + 2mg/L IBA were used. On the other hand, statistical differences were nil between all medium type as number of leaves was concerned. Meanwhile, table (2-C) reflects that the combination between Lloyd, G. and B.H. McCown²¹ medium free as well as Murashige and Skoog¹⁹ medium free and Black MissionFig cultivar were significantly improved number of roots in comparison with the other combination.

Table (2): Effect of different medium type on growth parameter during rooting stage of two Fig cultivars.**Table (2-a): Effect of cultivar:-**

Growth parameter Cultivar	Shoot length (Cm)	Number of roots (No.)	Root length (Cm)	Number of leaves (No.)
BlackMission	5.78a	12.08a	2.74	6.44
Conadria	3.49b	3.91b	2.11	5.60
L.S.D	1.02	1.86	NS	NS

Means followed by the same letter(s) with each parameter are not statistically different 5% level.

Table (2-b): Effect of medium type:-

Growth parameter Medium type	Shoot length (Cm)	Number of roots (No.)	Root length (Cm)	Number of leaves (No.)
Free MS	4.89ab	9.55ab	1.92bc	6.50
Free WP	5.78a	12.06a	4.45a	6.72
MS+2mg/L IBA	3.57b	1.84c	0.63c	4.64
WP+2mg/L IBA	5.19a	8.54b	2.70ab	6.22
L.S.D	1.44	2.64	1.78	NS

Means followed by the same letter(s) with each parameter are not statistically different 5% level.

On contrast, different combination investigated failed to induce statistical differences with shoot length, root length and number of leaves.

In general, the above mentioned resulted conclude that BlackMission cultivar recorded the highest number of roots and shoot length as compared with Conadria cultivar. Meanwhile, highest number of roots (12.06) and root length (2.45) was observed in woody plant medium free. The above mentioned results assured the findings of²⁶. They reported that the used of different concentrations of NAA or IBA had no significant effect on raising rooting parameters as compared with auxin free medium of Fig (*Ficus caracal*). On the other hand, there are reports of rooting in Fig cultivars at varying level of 1.0 mg/L IBA²⁷, 1mg/L⁵ and hormone free medium².

Table (2-C): Effect the interaction between cultivar and medium type

Growth parameter Cultivar	Shoot length (Cm)		Number of roots (No.)		Root length (Cm)		Number of leaves (No.)	
	Black Mission	Conadria	Black Mission	Conadria	Black Mission	Conadria	Black Mission	Conadria
Free MS	5.67	4.11	16.33	2.76	2.72	1.11	6.44	6.56
Free WP	7.06	2.50	18.00	6.12	4.57	4.33	7.11	6.33
MS + 2mg/L IBA	3.72	3.42	1.67	2.01	0.25	1.01	5.11	4.17
WP + 2mg/L IBA	6.67	3.72	12.33	4.75	3.41	1.98	7.11	5.33
L.S.D	NS		5.53		NS		NS	

Means followed by the same letter(s) with each parameters are not statistically different 5% level.



Figure 1: showed growth performance of three *Ficuscarcia* cultivars (Con: Conadria, B: Black Mission) meanwhile numbers indicates to medium type

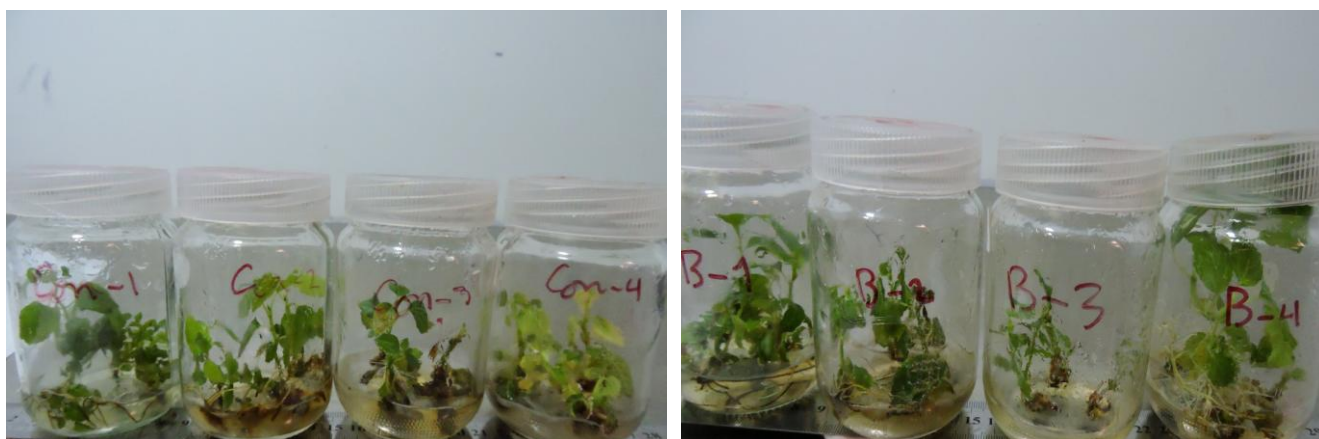


Figure 2: showed growth performance of three *Ficuscarcia* cultivars (Con: Conadria, B: Black Mission) meanwhile numbers indicate to medium type.

Acknowledgement:

We would like to express our grateful for our institute "National Research Centre , Cairo, Egypt" to support this work.

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