

Evaluate the possibility of producing citrus grafted plant simultaneously rooting cuttings

Iman Mosalanejad, Abdolhossein Aboutalebi* and Behnam Behrooznam

Department of Horticulture, Jahrom Branch, Islamic Azad University, Jahrom, Iran

Corresponding author: Abdolhossein Aboutalebi

ABSTRACT: In order to evaluate the possibility of producing citrus grafted plant simultaneously rooting cuttings, was performed an experiment as factorial arrangement in completely randomized design by using Bakraei (*C. reticulata* × *C. limmetta*) as rootstock and Sweet lime (*C. limmetta*) as scion with four replications. The 15-cm-cuttings were prepared from annual shoots of Bakraei and were budded by Sweet lime scion. The treatments for scion growth simulation were consisting GA₃ and BA 50 mgL⁻¹, which was applied before budding operation. The budded cuttings were treated by IBA 0, 1000, 2000 and 3000 mgL⁻¹ before plantation and then were cultured in the pots containing peat-moss and were transferred to the greenhouse equipped by mist system. The traits were measured after 120 days. Based on the obtained results, IBA had significant influence on rooting and the highest rooting percent (54.2%) obtained from IBA 3000 mgL⁻¹ treatment. The effect of BA on graft union percent and scion length was significant than GA₃. Generally, in order to production of Sweet lime grafted plant simultaneously rooting of Bakraei cutting can be used IBA 3000 mgL⁻¹ as rooting simulator and BA 50 mgL⁻¹ as graft union simulator.

Keywords: Budding, Cutting, Grafted plant, IBA, BA, GA₃.

INTRODUCTION

Citrus is one of the most important horticultural crops, which its cultivation in Iran back to the year round. According to the segregation in the citrus seedlings, the importance of grafting and production of uniform plants is clear. Selection of suitable rootstock and scion lead to increasing quality, more yielding and early ripening, which must be performed many researches to determine the best rootstock and scion for one of the cultivars. Regards to the need time for seedling growth until grafting operation, It seems using of cutting for rootstock production be the suitable option to reduction the time of rootstock production and prevention of segregation. Due to various aspects of this method can be used from different helpful rhizogenesis and graft union materials till graft union and root production time is minimized. The most common citrus propagation method in Iran is usage of the grafted plants. Grafting method has many advantages such as shortening juvenility period and production of uniform trees from a suitable cultivar. In this method the first seed of rootstock (Mexican lime, Sour orange, Bakraei etc.) is planted and after 6-12 months and preparing the plant to grafting, different citrus cultivars are budded on rootstock. In this method usually is used inverted T-budding that its graft union percent is high. The grafted plants are kept about one year in dense plots and then are transferred to the main field with some soil of around root (Fotuhi-Ghazvini, 1999 and Khosh-khui, 1989). In this study was tried according to segregation of seedlings and long-term period to prepare of seedling for grafting be replaced an alternative method. In this regards, the done researches are very low and it seems in the future can be developed application of clone rootstocks such as cutting. Different groups of plant growth regulators such as Auxins, Gibberellins, Cytokinins, Ethylene, Bracinoids and phenolic monologues are effective on root initiation. In between, Auxins have the most important effect on rooting of the cuttings. Some of the plant growth regulators are exciter and intensifier of roots growth and some of them are inhibitor of rot growth (Khosh-khui, 1989). Indol butyric acid (IBA) is one of the best and the most common materials for rooting. This material has little auxin effect and the parser enzymes slowly analyze it (Weaver, 1972). (Eshghi and Tafazoli, 2006) to evaluate the shortening period of Mexican lime seedlings growth for grafting and its union by application of

plant growth regulators and pinching in Shiraz university found that all treatments in comparison with control treatment significantly increased length and diameter and fresh weight of the shoots but application of GA₃ alone increased internode length than control treatment and Ethephon and Paclobutrazol decreased graft union percent. Generally, grafting simultaneously planting of the cuttings that named table-grafting is a new grafting method in the world and has been performed only on some plants. (Fontanazza and Rugini, 1983) to evaluate the histology of graft location in propagation of Olive trees by graft-cutting method and replacing this method to graft on seedling, selected 'DA-12-1' rootstock (an easy-rooting cultivar) and 'Giarafa' scion (a hard-rooting cultivar) and after grafting by splice-grafting method and placing grafted-cuttings in the boxes containing suitable medium and transferring the boxes to greenhouse equipped by bottom heat and mist system and temperature of 25 °C observed that in a short duration, meanwhile rooting of the cuttings, vascular connection between rootstock and scion continued after two months. (Mergre et al., 2007) performed table-grafting for two species of Rhododendron and distinctly recommended this propagation method as a fast propagation technique for the species that have weak rooting ability. (Bordbar, 2009) to assay the possibility production of the grafted plants from Olive cutting during rooting duration, grafted three scion cultivars consist 'Fishmi', 'Shiraz' and 'Dezful' on two olive rootstocks consist 'Shengeh' and 'Kargan' in two times consist simultaneously cutting planting and graft on the rooted cuttings. His results indicated that graft on the rooted cuttings had more graft union percent and long-term duration. Thus inverted-T-budding method was better than splice grafting method and duration of grafted plant in the first time grafting (simultaneously cutting planting) was 90 days. The aims of the present study were production of the grafted citrus trees in a short duration, better quality and reducing of production costs.

MATERIALS AND METHODS

In order to evaluate the possibility of producing citrus grafted plant simultaneously rooting cuttings, was performed an experiment as factorial arrangement in completely randomized design by using Bakraei (*C. reticulata* × *C. limmetta*) as rootstock and Sweet lime (*C. limmetta*) as scion and Gibberellic acid (GA₃), Benzyl Adenine (BA) and Indolebutyric acid (IBA) with four replications in Jahrom township. The first factor was rooting treatment (IBA 0, 1000, 2000 and 3000 mgL⁻¹) and the second factor was scion growth simulator (GA₃ and BA 50 mgL⁻¹). The 15-cm-cuttings were prepared from annual shoots of Bakraei in the 6 November from the trees of garden in the Pars-Narang Company. The cuttings had maximum two terminal leaves. The scions were prepared from 5-6 years old Sweet lime trees. The treatments were consisting: T₁) Control (cuttings without rooting treatment and scion); T₂, T₃, T₄) Cuttings without rooting treatment grafted by without treatment scion or by the treated scion with BA and GA₃ 50 mgL⁻¹ treatments respectively; T₅, T₆, T₇) Treated cuttings by 1000, 2000 and 3000 mgL⁻¹ IBA grafted by the without treatment scion respectively; T₈, T₉, T₁₀) Treated cuttings by 1000, 2000 and 3000 mgL⁻¹ IBA grafted by the treated scion with 50 mgL⁻¹ BA respectively; T₁₁, T₁₂, T₁₃) Treated cuttings by 1000, 2000 and 3000 mgL⁻¹ IBA grafted by the treated scion with 50 mgL⁻¹ GA₃ respectively. The supplied scions were dipped in the GA₃ or BA treatments for 20 seconds and then were budded on the cuttings. Graft location was closed by thin and white plastic bands. Then the cuttings were dipped in different concentration of IBA based on the treatment type for 20 seconds. The cuttings were cultured immediately after above operations in the pots containing peat-moss and were transferred to the greenhouse equipped by mist system. The traits such as the grown scions percent, scion length, the rooted cuttings percent and root fresh weight were measured after 120 days. Statistical analysis was done by using MSTAT-C software and the means were compared by Duncan's multiple range test (DMRT).

RESULTS AND DISCUSSION

Based on the results of analysis of variance table, rooting simulator treatment by application of different concentrations of IBA had significant influence ($p < 0.01$) on root fresh weight, rooting percent and scion length but had no significant effect on the grown scions percent. Effect of scion growth simulator treatments were significant ($p < 0.01$) on the grown scions percent, scion length and root fresh weight but had no significant influence on rooting percent. There was significant interaction ($p < 0.05$) between rooting and scion growth simulators only in relation to root fresh weight (Table 1).

Table 1. Analysis of variance (Mean Square) in relation to the evaluated characteristics

S.V	D.F	Mean Square			
		Grown cuttings percent	Scion length	Rooting percent	Root fresh weight
Rooting simulator (A)	3	324.5 ^{ns}	0.345 ^{**}	3547.3 ^{**}	0.0205 ^{**}
Scion growth simulator (B)	2	10855.6 ^{**}	3.988 ^{**}	92.7 ^{ns}	0.0059 ^{**}
Interaction AB	6	68.7 ^{ns}	0.061 ^{ns}	72.8 ^{ns}	0.0010
Error	36	198.2	0.040	54.8	0.0003
C.V		39.6	27.4	23.9	20.4

^{ns} not significant; ^{*}, ^{**} significant in 5 and 1% respectively

Effect of different concentration of IBA (rooting simulator) on the evaluated characteristics

IBA had no significant influence on the grown scions percent but scion length increased by increasing the concentration of IBA so that there was significant difference between control and IBA 3000 mgL⁻¹ treatments in relation to scion length. Rotting percent in control treatment was 14.2%, which increased by using IBA. The highest rooting percent (54.2%) was relative to IBA 3000 mgL⁻¹. Thus there was significant difference (p<0.01) between various levels of IBA in relation to root fresh weight (Table 2).

Table 2. Effect of different concentrations of IBA as rooting simulator on evaluated traits

Traits	IBA concentrations			
	0	1000	2000	3000
Grown cuttings percent	†34.4 ^a	32.1 ^a	32.4 ^a	43.2 ^a
Scion length (cm)	0.57 ^b	0.79 ^{ab}	0.62 ^b	0.94 ^a
Rooting percent	14.2 ^d	23.0 ^c	32.7 ^b	54.2 ^a
Root fresh weight (g)	0.043 ^d	0.073 ^c	0.094 ^b	0.142 ^a

†Means in each row having the same letter, have not significant difference (P ≤ 0.01) according to DMRT

The obtained results almost is according to the findings of (Ozelbaykal and Gezerel, 2005), which among concentration of 0-6000 mgL⁻¹ IBA for rooting of olive cuttings recommended 6000 mgL⁻¹ as the best concentration. (Aboutalebi et al., 1996) also reported the best range of IBA concentration 2000-3000 mgL⁻¹ for rooting of olive cuttings that was conformed to the results of present study. (Attarzadeh, 2009) reported that the best level of IBA in order to rooting of olive cuttings is 4000 mgL⁻¹ for 5 seconds accompanying Hydrogen Peroxide 3.5% for 30 seconds, which is according to the results of our study.

Effect of scion growth simulator (BA and GA₃) on the evaluated characteristics

BA and GA₃ treatments had no significant influence on root fresh weight and rooting percent but the effect of these treatments were significant on scion length and the grown scions percent in comparison with control treatment. The greatest percent of the grown scions (65.4%) was relative to BA treatment and GA₃ and control treatments were in the same statistical class. Scion length also was in the highest level (1.26 cm) in BA treatment and GA₃ and control treatments were in the next rankings (Table 3).

Table 3. Effect of scion growth simulators on evaluated traits

Traits	treatments		
	Control	BA 50	GA3 50
Grown cuttings percent	†17.9 ^b	65.4 ^a	23.2 ^b
Scion length (cm)	0.26 ^c	1.26 ^a	0.68 ^b
Rooting percent	28.6 ^a	31.1 ^a	33.4 ^a
Root fresh weight (g)	0.108 ^a	0.070 ^b	0.086 ^b

†Means in each row having the same letter, have not significant difference (P ≤ 0.01) according to DMRT

Effect of the derived treatments from interaction between rooting and scion growth simulator on the evaluated characteristics

The highest scion growth percent was relative to IBA 3000 mgL⁻¹ and BA 50 mgL⁻¹ (75.8%). This treatment had no significant difference with BA 50 mgL⁻¹ in each IBA levels. In fact, auxin treatment did not influence on scion growth and the main effect was relative to BA treatment. The lowest scion growth percent was observed in control treatment (16.5%). In this relation, the treatments containing GA₃ or without scion growth simulator in all IBA levels had no significant difference with control treatment. This result indicated that GA₃ and IBA are not effective on scion growth (Table 4).

The greatest scion length was relative to IBA 1000 and 3000 mgL⁻¹ and BA 50 mgL⁻¹ (1.40 and 1.38 cm respectively). IBA treatments had no influence on scion length and did not observed any enhancement in the scion length by increasing IBA concentration but BA had significant difference to others in all concentrations of IBA, which indicated significant effect of BA on scion length (Table 4).

The highest rooting percent was relative to IBA 3000 mgL⁻¹. BA and GA3 treatments had no influence on rooting percent and there was no significant difference between application of these treatments and non-application of them. Rooting percent significantly increased by increasing IBA concentration.

The greatest root fresh weight was observed in IBA 3000 mgL⁻¹ and non-application of scion growth simulator treatments. Scion growth treatments had no influence on root fresh weight. In contrast, rooting hormone increased root fresh weight by increasing concentration. Application of scion growth simulator approximately had inversed effect on root fresh weight and led to reduction of root fresh weight but increasing IBA concentration increased root fresh weight.

Table 4. Effect of the derived treatments from interaction between rooting and scion growth simulator on the evaluated characteristics

Traits		Grown cuttings percent	Scion length (cm)	Rooting percent	Root fresh weight (g)
IBA × Scion growth simulator					
IBA 0	without treatment	16.5 ^b	0.08 ^f	13.8 ^{ef}	0.040 ^f
	BA 50 mgL ⁻¹	66.2 ^a	1.00 ^b	10.0 ^f	0.038 ^f
	GA ₃ 50 mgL ⁻¹	20.5 ^b	0.62 ^{cd}	18.8 ^{def}	0.052 ^{ef}
IBA 1000 mgL ⁻¹	without treatment	20.0 ^b	0.32 ^{def}	20.8 ^{cdef}	0.092 ^{cd}
	BA 50 mgL ⁻¹	57.8 ^a	1.40 ^a	25.8 ^{cd}	0.062 ^{ef}
	GA ₃ 50 mgL ⁻¹	18.5 ^b	0.65 ^c	22.5 ^{cde}	0.065 ^{def}
IBA 2000 mgL ⁻¹	without treatment	15.0 ^b	0.20 ^{ef}	25.8 ^{cd}	0.120 ^{bc}
	BA 50 mgL ⁻¹	62.0 ^a	1.25 ^{ab}	32.5 ^{bc}	0.070 ^{de}
	GA ₃ 50 mgL ⁻¹	20.2 ^b	0.42 ^{cde}	39.8 ^b	0.092 ^{cd}
IBA 3000 mgL ⁻¹	without treatment	20.0 ^b	0.45 ^{cde}	54.0 ^a	0.180 ^a
	BA 50 mgL ⁻¹	75.8 ^a	1.38 ^a	56.0 ^a	0.110 ^{bc}
	GA ₃ 50 mgL ⁻¹	33.8 ^b	1.00 ^b	52.5 ^a	0.135 ^b

^fMeans in each column having the same letter, have not significant difference (P ≤ 0.01) according to DMRT

CONCLUSION

Application of IBA is recommending for rooting of Bakraei cuttings and the best IBA concentration is 3000 mgL⁻¹. The best treatment for graft union and scion growth was BA 50 mgL⁻¹. Generally, BA 50 mgL⁻¹ for scion growth and IBA 3000 mgL⁻¹ for rooting of the cuttings had better results in comparison with all the used treatments.

Recommendations

It recommends that this study must be performed in various regions to achieve the best grafting time. Thus other concentrations of BA such as 100-200 mgL⁻¹ and IBA such as 4000 mgL⁻¹ must be used. It recommends that a study must be conducted in this relation and the produced plants culture in the main field.

REFERENCES

- Aboutalebi A. 1996. Evaluation the effects of cutting time, cutting position on the shoot, rooting medium and auxins hormones on rooting of the Olive c.v 'Zard'. M.Sc Thesis, Islamic Azad University of Jahrom. (In Persian)
- Attarzadeh M. 2009. Evaluation the effect of hormonal treatments and rooting helper materials on rooting of the hard-rooting cultivars of Olive. M.Sc Thesis, Islamic Azad University of Jahrom. (In Persian)
- Bordbar GH. 2009. Evaluation the possibility production of the grafted plants from Olive cuttings in rooting time. M.Sc Thesis, Islamic Azad University of Jahrom. (In Persian)
- Eshghi S, Tafazoli E. 2006. Reduction the growth period of Mexican lime seedlings for grafting and graft union by using plant growth regulators and pinching. Journal of Agricultural and Natural Resources Sciences and Techniques, Vol. 4. (In Persian).
- Fontanazza G, Rugini R. 1983. Graft union histology in Olive tree propagation by cutting-grafts. Riv. Ortoflorofrut, IT. SV.
- Fotuhi-Ghazvini R. 1999. Citrus growing in Iran. Guilan University Press. p. 2-6.
- Khosh-khui M. 1989. Plant propagation, principles and practices. Shiraz University Press. 1586 p. (In Persian)
- Mergre D, Kondratovics U, Dakane K. 2007. Simultaneous graft union and adventitious root formation during vegetative propagation in *Elepidote rhododendrous*. Acta Universitatis Latviensis. Vol. 723, Biology, pp. 155-160.
- Weaver R J. 1972. Plant growth substances. W.H. Freeman and Company, 594 p.