

The Effects of Cutting Collection Time, Auxin Types and Auxin Concentrations on Rooting of Olive *Olea europaea* L. Cuttings Under Arid Land Conditions.

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Abstract

The effect of different concentrations of IBA and NAA at 0, 1000, 3000, 6000 and 9000 ppm on rooting of olive cuttings collected at three different seasons (autumn, winter and spring) were examined under arid land conditions. Cuttings collected in autumn (November) and winter (January) showed similar responses for percentage of rooting (39.33 & 44.0 %), number of roots (5.32 & 5.93 roots) and root length (4.42 & 5.04 cm) respectively. However, cuttings collected later on spring (March) had statistically lower rooting percentage (19.66 %), root number (19.66 roots) and root length (3.16 cm). When using IBA, higher percentage of rooting (43.55%), root number (6.06 roots) and root length (5.29 cm) were detected as comparing with the NAA treatments (25.11%), (3.38 roots) and 3.11 cm), respectively. Data obtained under the effects of auxin concentrations showed that treatment with 3000 and 6000 ppm had the highest percentage of rooting (48.88 & 46.11 %) and root length (4.59 & 5.44 cm), while other concentrations exhibited lower responses.

Key words: Stem cutting, Auxin, IBA, NAA, Season, Olive.

1. Introduction

Olive *Olea europaea* L. is an important native plant in arid and semiarid region of the Mediterranean region, central Asia and various part of Africa [12]. Olive can be vegetatively propagated by grafting or budding on seedling rootstock [8]. However, vegetative propagation of Olive by stem cuttings is considered to be an easy, inexpensive and appropriate for mass plant production [7]. Low rooting ability of Olive can be the result of not choosing the best time of removing cuttings from the mother plants [8] and cultivars [14]. Plant growth regulators can play a vital role to enhance rooting of Olive cuttings [1]. However, Indole-3-butyric acid (IBA) and Naphthalene acetic acid (NAA) increase rooting responses of Olive stem cuttings cultivars [3]. Progeny of Olive trees has been successfully grown at King Abdulaziz University Agric. Research station orchard for more than 20 years ago as an ornamental plantation. No propagation techniques have been established for those adapted Olive trees to local environment, soil and water salinity. The study was, therefore, carried out to examine the effect of different concentrations of IBA and NAA on rooting of olive cuttings collected at three different seasons; late autumn (mid- November); mid-winter (January) and early spring (mid-March).

2. Materials and Methods

2.1. Experimental site and treatments

The study was carried out during late autumn (mid- November), winter (mid-January) and spring (mid-March) of 2011 and 2012 at the propagation units of King Abdulaziz University (KAU) Agricultural Research Station nursery at Hada Al-Sham, Jeddah, Saudi Arabia . Semi-hard wood cuttings of Olive *Olea europaea* L. were obtained from existing progeny in an orchard of KAU Agricultural Experiment station, which were planted for more than 20 years. The trees were severely pruned for the last 3 years of the experiment in order to stimulate the production of juvenile vegetative shoots. 15- 20 cm of semi hard wood cutting with 4-6 leaves were collected at three different seasons autumn (mid- November), winter (mid-January) and spring (mid-March). The base of each cutting was treated for

5 seconds with the different PGR treatments dissolved in a 45% solution of ethanol while the control treatment was performed with dipped in a 45% ethanol solution only. Indole-3- butyric acid solution (IBA) and Naphthalene acetic acid (NAA) at 0, 1000, 3000, 6000 and 9000 ppm were investigated. 2-3 cm of each cutting was dip with the different treatments and immediately planted in poly ethylene tube (size 10) filled with 90% perlite and 10% peat moss. The treated cuttings were kept in controlled environment greenhouse (25-28 C; humidity 65-80%; lighting 7000 -10000lux) and humidity of treated cuttings was increased manually by hand sprinkling three times in a day. Three replicate were used for each treatment and ten cuttings were planted in each replicate. Thirty cuttings were used for each PGR treatment with total of 90 cuttings for each collection time (season). Cuttings were removed from the poly ethylene propagation tube in 90 days and data were recorded on percentage of rooted cuttings, number of root per cutting and root length for each GPR treatment and season.

2.2. Statistical Design and Analysis

The experiment was arranged as split- split plot design with 3 replicates of 10 cuttings for each replicate. Main plot treatment being the three different collection time of stem cuttings (Autumn, Winter and Spring). The sub plot treatment was the auxins (IBA and NAA). Sub-sub plot treatments being the five auxin concentrations (0 ,1000, 3000,6000 and 9000 ppm). Data for rooting percentage (%), root number and root length(cm) were statistically analyzed using analysis of variance method. Means were statistically compared using the LSD test at $p \leq 0.05$. The statistical analysis was performed by using [13] according to [6].

3. Results

3.1. Analysis of variance

Overall results showed significant differences at $p \leq 0.01$ among cutting collection time, auxin type and auxin concentrations for all treaties under investigation (rooting percentage, root number and root length). The interaction between cutting collection time and auxin type was significant at the $p \leq 0.05$ on root number, but was not significant for both rooting percentage and root length. Significant effects were showed at $p \leq 0.01$ for the effect of the interaction between cutting collection time and auxin concentrations on the 3 studied traits. The interaction among auxin types and auxin concentration showed significant results at $p \leq 0.05$ on root number and root length, but significant observation was detected at the 5% level for rooting percentage. However, there were also different responses were detected among the interaction of cutting collection time, auxin type and auxin concentrations, but the effects observed were not significant (Table 1).

Table(1): Analysis of variance for percentage of rooting, root number and root length of *Olea europaea* L. for cutting collection time, auxin type, auxin concentration and their interactions.

Sources of variation	df	Percentage of rooting	Number of roots	Root length
Replications	2	423.33 ns	6.45 ns	2.12 ns
Cutting collection time(T)	2	5003.33 **	77.17 **	27.66 **
Error a	4	26.66	1.85	1.97
Auxin type(A)	1	7654.44 **	160.72 **	107.1 **
TxA	2	414.44 ns	9.29 *	1.23 ns
Error b	6	401.11	3.62	4.35
Auxin concentrations(C)	4	2762.22 **	97.91 **	24.38 **
TxC	8	88.05 ns	8.35**	1.58 ns
AxC	4	565.55 *	27.755 **	15.7 **
TxAxC	8	54.72 ns	2.73 ns	0.74ns
Error c	48	210.55	2.39	1.94

ns, not significant at $p \leq 0.05$. *, **, significant at $p \leq 0.05$ and $P \leq 0.01$, respectively. •

3.2. Effect of cutting collection time

As shown in Table 2, cuttings collected in winter (January) produced the highest responses for percentage of rooting (44.0 %), number of roots (5.93 roots) and root length (5.04 cm) respectively. However, cuttings collected later on spring (March) had statistically lower rooting percentage (19.66 %), root number (19.66 roots) and root length (3.16 cm).

Table(2): Means of percentage of rooting, root number and root length of *Olea europaea* L. under the effect of different cutting collection time.

Cutting collection time	Percentage of rooting(%)	Root number	Root length(cm)
Autumn (November)	39.33 b	5.32 a	4.42 a
Winter (January)	44 a	5.93 a	5.04 a
Spring (March)	19.66 c	2.9 b	3.16 b

* Means followed by the same letter are not significantly different according to LSD at $P \leq 0.05$

3.3. Effect of auxin type

Treatment with different auxin type showed statistically different responses. When using IBA, higher percentage of rooting (43.55%), root number (6.06 roots) and root length (5.29 cm) were detected as comparing with the NAA treatments (25.11%), (3.38 roots) and 3.11 cm), respectively (Table 3).

Table(3): Means of percentage of rooting(%), root number and root length(cm) of *Olea europaea* L. under the effect of different axin types .

Auxin type	Percentage of rooting(%)	Root number	Root length(cm)
NAA	25.11 b	3.38 b	3.11 b
IBA	43.55 a	6.06 a	5.29 a

* Means followed by the same later are not significantly different according to LSD at $P \leq 0.05$

3.4. Effect of auxin concentration

Data obtained under the effects of auxin concentrations showed that treatment with 3000 and 6000 ppm had the highest percentage of rooting (48.88 & 46.11 % respectively) while the other concentration exhibited lower and statistically similar percentage of rooting (table 4). Data obtained from root number showed that treatment with the auxin concentration 3000 ppm had the highest number of roots (8.14 roots). Treatment with 6000 ppm exhibited lower root number (5.92 roots) compared to the 3000 ppm treatment. Fewer root number was observed with other auxin concentration and were statistically similar (Table 4). Under the effect of 6000 ppm longer roots (5.44 cm) and was showed statistically similar to the 3000 ppm treatment (4.94 cm). Treatment with 1000, 3000 and 9000 ppm showed no significant differences in root length, but longer root were observed when compared to the control treatment (2.38 cm) as shown in Table 4.

Table(4): Means of percentage of rooting(%), root number and root length(cm) of *Olea europaea* L. under the effect of different auxin concentrations .

Auxin concentration	Percentage of rooting(%)	Root number	Root length(cm)
0 ppm (control)	22.22 b*	2.24d	2.38 c
1000 ppm	30 b	3.94 c	4.10 b
3000 ppm	48.89 a	8.14 a	4.95 ab
6000 ppm	46.11 a	5.92 b	5.44 a
9000 ppm	24.44 b	3.37 c	4.15 b

* Means followed by the same later are not significantly different according to LSD at $P \leq 0.05$

3.5. Effect of the interaction between cutting collection time and auxin concentrations

The interaction between cutting collection times and auxin concentrations (Table 5) showed significant differences at $p \leq 0.01$ on root number. However, data obtained from percentage of rooting and root length showed different responses, but they were not statically different. Collecting cuttings in autumn or winter showed the highest root number when treated with 3000 ppm than cutting collected later on the season (spring). Fewer root number were obtained with the control and 9000 ppm auxin concentrations when collecting cuttings in spring and autumn seasons.

Table(5): Means of root number of *Olea europaea* L. under the effect of the interaction between cutting collection times and auxin concentrations.

Cutting collection time (date)	Auxin concentration(ppm)	Root number
Autumn	control	2.12
	1000	4.08
	3000	10.17
	6000	6.47
	9000	3.74
Winter	cotrol	3.41
	1000	5.00
	3000	10.29
	6000	6.87
	9000	6.36
Spring	control	1.17
	1000	2.72
	3000	3.95
	6000	4.41
	9000	2.24
LSD (0.05)		1.97

3.6. Effect of the interaction between auxin type and auxin concentrations

The interaction between auxin types and auxin concentrations showed significant interaction at $p \leq 0.01$ level on root number and length and at the $p \leq 0.05$ level on percentage of rooting. When using IBA at 3000 ppm showed higher rooting percentage compared to other treatments. The control treatment, 1000 and 9000 ppm showed fewer root number with NAA. Similar statistically responses were observed when using NAA or IBA at control, 1000 and 9000 ppm. However, treatment with IBA at 3000 ppm exhibited the highest root number overall. Root length showed statistically similar responses when using NAA at control, 1000 and 3000 ppm. Higher concentrations of NAA (6000 and 9000 ppm) showed the highest root length under the NAA concentrations and were statically similar. However, when using IBA similar statistically responses were observed on root length at all concentrations except the 6000 ppm. The treatment of IBA with 3000 ppm produced the highest rooting (%) root number and root length compared with the other NAA or IBA treatments as shown in table 6.

Table(6): Means of percentage of rooting(%), root number and root length(cm) of *Olea europaea* L. under the effect of the interaction between auxin types and auxin concentrations.

Auxin types	Auxin concentrations (ppm)	Rooting percentage (%)	Root number	Root length (cm)
NAA	control	21.11	2.28	2.51
	1000	21.11	2.45	3.10
	3000	32.22	4.86	3.86
	6000	37.77	5.27	4.44
	9000	13.33	2.04	1.65
IBA	control	23.33	2.19	2.25
	1000	38.88	5.42	5.10
	3000	65.55	11.41	6.03
	6000	54.44	6.57	6.43
	9000	35.55	4.69	6.65
LSD (0.05)		13.75	1.46	1.32

4. Discussion

Data showed that it is essential to choose the best time for removing cuttings from the mother plant. A cross different treatments of IBA and NAA, cuttings collected in autumn and winter showed higher rooting responses than the one collected later on the season (spring). This could be the result of seasonal change of carbohydrates in reproductive and vegetative shoots and the direct products of photosynthetic activity [3]. Thus, the finding of [4] and [10] suggests that in Olive cuttings soluble sugars are more important in root initiation than starch. In addition, starch concentration was lower than those of individual sugars. Furthermore, due to seasonal changes it appears

that the base of cutting has an important factor in root initiation due to the availability of carbohydrates [10]. Due to long rooting process of Olive cuttings and the very low rate of photosynthesis of Olive cuttings; therefor only two or three leaves should be left on Olive cuttings which promoted rooting when combined with IBA treatments [2]. The result showed that the treatment with the Auxin (IBA & NAA) has positively affected rooting of Olive cuttings. Since hormonal balance regulates the process of root formation and elongation, with auxin dominating the initiation of roots. In olives the natural supply of auxin from the apical buds of the cutting through the base is limited in most varieties, there is a need to supply exogenous auxin[14]. Exogenous supply of auxin with balance between endogenous stimulatory and inhibitory factors, in addition to, nutritional factors is required to promote rooting of Olive cuttings [15]. Application of IBA showed better responses on percentage of rooted cutting, root number, and root length when compared with NAA at all different seasonal collection time. Similar findings have been reported by [1] and [9]. However, [5], report showed that various concentrations of IBA and NAA on Olive cuttings has positive effect in overall rooting responses and concluded that an auxine treatment alone was not sufficient to stimulate meristematic activity and root initiation in certain cultivars. Percentage of rooted cuttings and root number decreases as IBA concentration increase. Similar findings has been reported by [2]. However, higher concentration of both auxin showed possitive effect on root length over the different collection seasons. The study of [15] explained that the higher number of root with IBA treatment s was due to the physiological interaction of IBA and carbohydrates.

5. Conclusions

The results of the present study suggest that IBA increases the rooting ability among the different cuttings collection times when compared to the NAA treatments. Autumn and winter period is the best season for rooting of Olive *Olea europaea* L. cuttings obtained from existing progeny in an orchard of KAU Agricultural Experiment station. The IBA with 3000 ppm was the highest treatment in rooting (%), root number and root length. Our results confirm previous reports that the availability of carbohydrates in the base of cuttings appears to be an important factor in root initiation.

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