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Effects of Different Auxin (IBA) Concentrations and Planting-Beds on Rooting Grape Cuttings (*Vitis vinifera*)

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Authors' contributions

This work was carried out in collaboration between all authors. Authors MG and MAK designed the study, performed the statistical analysis, wrote the protocol and managed the analyses of the study. Authors SRM and MG managed the literature searches and wrote the first draft of the manuscript. Author MG read and approved the final manuscript.

Research Article

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ABSTRACT

Aims: *Vitis vinifera* is a grape species and native to the Mediterranean region and east to northern Iran. The present research was carried out in greenhouse conditions to study the effects of four concentrations of indole-3-butyric acid (IBA) (0; 2000; 4000; and 6000 mg/l) and three planting beds (agricultural soils, sandy, and mixture of agricultural soils and sand) on rooting grape cuttings in institute of agriculture at Zabol University, (Iran).
Methodology: The experimental design was a factorial design in randomized complete block with three replicates.
Results: Results showed that different auxin and planting bed treatments had a significant influence on grape rooting. The maximum number of roots, root length, and root fresh and dry weight was obtained by applying 4000 mg/l IBA. The significant effect of planting bed treatments was found in studying traits, so that maximum number of roots, root length, and root fresh and dry weight was obtained in mixture of agricultural soils and sand planting beds. Studied traits significantly affected by an interaction effect of IBA and cuttings beds, so that maximum number of roots, root length, and root dry weight was obtained by using 2000 mg/l IBA + sandy planting bed, and maximum root fresh weight was obtained by

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using 4000 mg/l IBA + sandy planting bed.

Keywords: Planting-beds; indole-3-butyric acid; cuttings; rooting.

1. INTRODUCTION

Grape (*Vitis vinifera*) (Common Grape Vine) is a species of Vitis, native to the Mediterranean region, central Europe, and southwestern Asia, from Morocco and north Portugal to southern Germany and east to northern Iran. Grape has the widest scattered range between treasurer fruit trees in the world and approximately a half hectare of each sixty hectare planted land are dedicated to grape. Ruby grape (*Vitis vinifera*) is one of the most important horticultural crops and seedless grape species in the east region of Iran (Sistan region) [1]. Sistan ruby grape is firstly and premature, which this is a good score. This product is marketable first week of May. There is little fruit in the market and demand is high at this time. Sistan ruby grape has unique features such as flavor and apparent luster, which considered as an advantage and can be an effective role in improving economic and social situation in the region [1]. Using hardwood cutting, semi - hardwood cutting or softwood cutting, taking cuttings at the appropriate time of the annual growth period of maternal plants, suitable temperature and humidity in rooting cuttings environment, using the optimal planting bed, application of plant growth regulators and finding optimal concentrations of these substances, are important factors that should be considered carefully for increasing rooting the cuttings. Auxin is one of the most important hormones that use on the stem cuttings for accelerating the formation of adventitious roots [4]. The essential role of auxin has been documented on induction rooting and root formation. Auxin has an effect on speed and increase the percentage of rooting the cuttings. Plants produced natural auxin in shoots and young leaves, but synthetic auxin should be used for success rooting to prevent of cuttings death [2,3]. Hartmann et al., repeated application of indole-3-butyric acid (IBA) in 2500 to 4000 mg/l had impressive results in rooting the semi - hardwood cuttings in apple, plum and olive [4]. Al-Sagri and Alderson showed that application of auxin in 3000 to 4000 mg/l had a positive effect in increasing rooting the cuttings of woody ornamental shrubs such as roses and Chinese hollyhock [5]. Babashpour-Asl et al. in evaluating the effect of Indole-3-butyric Acid on the rooting ability of semi-hardwood Bougainvillea (*Bougainvillea glabra*) cuttings reported there was no significant difference between IBA treatment and control on rooting percentage [6]. There was significant difference between IBA treatment and control on the number of main roots per cutting. The best effect of different levels of IBA on the number of main roots per cutting (8.67 roots per cutting) was obtained at 2000 mg/l IBA. Advantage's use of the growth regulators is as follows: increasing rate of rooting; increasing the percentage of rooted cuttings; increasing the number and quality of roots per cuttings; and uniformly rooting [7]. Rooting capacity for stem cuttings will be determined by the interaction of hereditary factors in stem cells and the following factors: auxin level; leaves and buds on the cuttings; the amount of food available in the cuttings; stage of plant growth; location of stem elongation and a type of the cutting tissue [8].

2. MATERIALS AND METHODS

The study was conducted in the Agricultural Institute of Zabol University Sistan and Baluchistan Province, Iran (30º 54' N, 61º 41' E). The region has an arid climate with 481 m altitude from sea level. Annual mean precipitation and temperature are 63 mm and 23ºC, respectively. During the 2012 growing season (120 Day), the experiment was conducted out

in a factorial design in randomized complete block with three replicates in greenhouse conditions. The auxin hormone treatments were in four levels as: (0; 2000; 4000; and 6000 mg/l) and three planting beds treatments were as: agricultural soils, sandy, and mixture of agricultural soils and sand. Physical and chemical properties of agricultural soil are presented in Table 1.

Table 1. Soil analysis result for physical and chemical characteristics

Characteristic	Soil depth cm	Soil texture	OC* %	EC dS m^{-1}	pH	N %	K mg kg^{-1}	P mg kg^{-1}
Value	0-30	Sandy-loam	1.63	1.60	7.50	0.07	148	10.40

**OC: Organic carbon; EC: Electrical conductivity*

Sandy soil was without the elements and organic matter with pH=7. The used auxin hormone was manufactured in Sweden. Sodium hydroxide (NAOH) was used for making auxin soluble. For this purpose added distilled water to 40 g solid sodium hydroxide (NaOH) and shake it until dissolved completely, then bring final volume of solution up to 1000 ml with the addition of more distilled water. In the next step 10 to 30 cc of the solvent was separated, and separately, 2000, 4000 and 6000 mg/l was used of auxin hormone, then bring final volume of solution; the results obtained hormones were as follows: 2000; 4000; and 6000 mg/l. Dishes containing auxin hormone were maintained at 4ºC inside the refrigerator and used over time. 25 Grape cuttings were selected by annual branches 20 cm length and contained five seedlings from the gardens of the Agricultural Institute of Zabol University. Cutting places was done with 1 cm distance to the buds and angled, to surface contact of cuttings be more with the soil and hormone. For treated the cuttings approximately 2.5 cm from the bottom of them was placed in rooting solution in 5 seconds. Then cuttings were disinfected with Benomyl (50%) fungicide. After treatment, 3 planting bed were prepared in pots as follows: agricultural soils, sandy, and mixture of agricultural soils and sand, and 25 cuttings were planted in each pot on 30 January 2012. Greenhouse humidity and temperature averaged were 60% and 25ºC. Three buds of each cutting were placed in the planting bed and two buds out of the soil. The cuttings were taken out of the soil and root number, root length, fresh and dry weight in each treatment was determined on 30 May 2012. Root fresh and dry weight was measured with GR-200 analytical balance, and roots were oven dried at 74ºC for 24h. Data analysis was done by using SAS and MSTATC software. The ANOVA test was used to determine significant ($p \leq 0.01$) treatment effect and Duncan Multiple Range Test to determine significant differences between individual means.

3. RESULTS AND DISCUSSION

Results showed that, the number of roots, root length, and root fresh and dry weight significantly affected by main effect of growth regulators and types of planting bed (Table 2). Number of roots increased with the main effect of auxin and different planting-beds treatments, so that maximum number of roots per cutting was 8.27 and 7.93 that was obtained with 4000 mg/l auxin and agricultural soils + sand planting-beds respectively (Table 3 and 4). Mean comparisons showed that maximum (60.05 cm) and minimum (32.82 cm) root length was obtained by application of 4000 mg/l auxin and control respectively (Table 3). Also main effect of planting-bed treatments significantly increased root length, so that maximum (61.77 cm) and minimum (25.72 cm) was obtained from agricultural soils + sand and agricultural soils respectively (Table 4). Root fresh and dry weight significantly increased

by main effect of auxin and different planting-bed treatments, so that maximum values was obtained with 4000 mg/l auxin and agricultural soils + sand planting-beds respectively (Table 3 and 4). Number of roots, root length, and root fresh and dry weight significantly decreased with 6000 mg/l auxin compared with 4000 mg/l (Table 3).

Table 2. ANOVA of the effects of auxin (IBA) and planting-beds on rooting grape cuttings

SOV	df	MS			
		Number of root per cutting	Root length (cm)	Root fresh weight (g)	Root fresh dry (g)
Replication	2	22.76	1641.13	0.031	0.00081
Auxin	3	11.88**[*]	1231.73**	0.0135**	0.006**
Planting-beds	2	19.07**	3998.63**	0.033**	0.027**
Auxin × Planting-beds	6	8.04**	560.89**	0.0054**	0.0048**
Error	22	1.71	135.30	0.00037	0.00031
CV (%)		19.64	25.62	14	21.68

** ns= Non significant, ** = p < 0.01*

Table 3. Means comparison of effects of auxin (IBA) on rooting grape cuttings

Treat	Number of root per cutting	Root length (cm)	Root fresh weight (g)	Root dry weight (g)
Control	5.58b[*]	32.820c	0.0965c	0.0750b
2000 (mg/l)	6.52b	48.570b	0.1457b	0.0985a
4000 (mg/l)	8.27a	60.050a	0.1850a	0.1125a
6000 (mg/l)	6.25b	40.170bc	0.1150c	0.0685b

** Columns means followed by the same letter are not significantly different at 0.01 probability* level

Table 4. Means comparison of effects of different planting-beds on rooting grape cuttings, agricultural soils, sandy, and mixture of agricultural soils and sand

Treat	Number of root per cutting	Root length (cm)	Root fresh weight (g)	Root dry weight (g)
Agricultural soil	5.415c[*]	25.72c	0.109b	0.0647b
Sandy	6.625b	48.72b	0.101b	0.0490c
Agricultural soil + Sand	7.937a	61.77a	0.796a	0.1385a

** Columns means followed by the same letter are not significantly different at 0.01 probability level*

The interaction effect of auxin and different planting-bed treatments was significant on the measured traits (Table 2). Maximum number of roots per cutting (9.91), root length (77.45 cm), and root dry weight (0.208 g) was at 2000 mg/l auxin and sandy planting-beds; moreover maximum root fresh weight (0.269 g) was obtained with 4000 mg/l auxin and sandy planting-beds (Table 5). Farooqi et al. [9] conducted an experiment on *Rosa damascena* Mill and studied the effect of IBA. They found the increasing trend of rooting percentage, number of roots per cutting, length of the longest root (cm), thickness of the root (cm), fresh weight of root and dry weight of root with increasing concentration of IBA from 100 mg/l to 300 mg/l [9]. Carvalho et al. opined that treating stem cuttings of *stevia* with IAA and IBA promoted rooting and increased the number of roots [10]. Chalapathi et al. reported that cuttings treated with IBA 500 mg/l was found to be superior with respect to shoot length,

number of branches, number of leaves and root length, survival percentage and sprouting percentage [11]. He also stated that better rooting and sprouting of *stevia* cuttings can be obtained through prolonged dipping of *stevia* cuttings in 50 mg/l IBA solution.

Researchers believe that high concentrations of auxin can cause damage to the cutting base. Auxin can be effective to rooting cuttings in a certain concentration, depending on the crop and cultivar, and will have an inhibition effect at higher concentrations [12]. The first root cell division and displacement movable rhizokalin to the rooting zone and their activation are performed in the presence of auxin in the root zone [13]. Based on results of present study number of roots, root length, root fresh and dry weight increased with increasing concentrations of IBA hormone from 2000 to 4000 mg/l compared to control; however, a concentration of 6000 mg/l had inverted effects. Alizadeh and Grigorian (2002) reported that, the increase in NAA hormone concentration from 1000 to 2000 mg/l increased rooting in semi hardwood cuttings of almond - peach hybrid, whereas decreased in rooting was observed at concentrations 3000 mg/l [14].

Table 5. Interaction effects of auxin and different planting-beds treatments on the measured traits

Treat	Number of root per cutting	Root length (cm)	Root fresh weight (g)	Root dry weight (g)
S1A1	4.00ef*	19.95c	0.079e	0.029e
S1A2	5.91cdef	34.32bc	0.091de	0.067bcd
S1A3	6.83cd	44.170b	0.118bcd	0.086b
S1A4	3.85f	19.37c	0.103cde	0.083b
S2A1	6.08cde	48.92b	0.0727e	0.078bc
S2A2	9.91a	77.45a	0.260a	0.208a
S2A3	9.41ab	75.05a	0.269a	0.183a
S2A4	8.08abc	73.30a	0.150b	0.057bcde
S3A1	7.33bcd	127.30bc	0.135bc	0.052bcde
S3A2	6.75cd	31.820bc	0.117bcd	0.046cde
S3A3	5.08def	38.37bc	0.092de	0.040de
S3A4	6.91cd	50.42b	0.0135bc	0.075bc

** Columns means followed by the same letter are not significantly different at 0.01 probability Level*

A_1 = Control (without auxin)
A_2 = 2000 (mg/l)
A_3 = 4000 (mg/l)
A_4 = 6000 (mg/l)

S_1 = Agricultural soil
S_2 = Sandy soil
S_3 = Agricultural soil + Sand

Application of synthesized auxin in high concentration can prevent of the bud development on the stem cuttings [15]. Also in the McGuire et al. research, the highest root length in oleander cuttings was obtained with 2000 and 3000 mg/l NAA, and 4000 mg/l showed negative effects on the rooting cuttings [16]. Abuo-Hadid in the studies stimulate rooting of cucumber cuttings with NAA hormone showed that, increased concentration from 500 to 1000 mg/l increased the number of roots; whereas concentration of 2000 mg/l had a deterrent effect on the radicle length [17]. The influence of different planting bed on the rooting of grape showed that rooting in mixture of agricultural soil + sand was better than in other treatments. The sand bed is poor of the nutrient and agricultural soil bed is facing with shortage ventilation; as a result combining these two beds can have a positive effect on rooting. Roots beginning were not significantly influence to the physicochemical properties of

beds, but the beds had an influence on the root elongation. Chen et al. in the reviews rooting of cuttings several ornamental plants showed that, adding compost to sphagnum peat or pine bark can be effective in rooting cuttings of plants [18].

4. CONCLUSION

In conclusion, current study results have proved the hypothesis that use of auxin could be a useful on rooting grape cuttings in the greenhouse condition. In the current research root fresh and dry weight, number of roots and root length significantly affected by growth regulators and types of planting bed. Agricultural soils as planting bed had a positive effect on rooting grape cuttings.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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