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EFFECT OF ROOTING HORMONES ON THE ROOTING CAPABILITY OF *Ficus benjamina* L. CUTTINGS

UTJECAJ FITOHORMONA NA SPOSOBNOST ZAKORJENJIVANJA REZNICA VRSTE *Ficus benjamina* L.

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Summary

Vegetative production techniques, of which cutting method widely used in propagation of ornamental plants, have a crucial role for conserving the plant genetic sources. On rooting development of stem cuttings, cutting position, rooting medium and rooting hormone are some of the critical factors that affect the success. The primary objective of this study is to determine the best hormone doses and ideal rooting medium on *Ficus benjamina* L. stem cuttings. Conventionally, the most frequently vegetative propagation method is the rooting of the stem cuttings in various media such as, pearlite, peat, sand, through exposure to high-concentration rooting hormones (IBA, IAA, NAA, etc.). But, this conventional technique requires wide areas in the rooting stage of mass production, prevents monitoring the course of rooting, and necessitates large amount of materials used as hormones and rooting media. In this study, a new method that may be preferable in mass production of plants was tested. 39 different treatments were carried out, and their results were evaluated. Sand, and perlite were used as solid rooting media. Stem cuttings were kept in low-concentration hormones permanently after cutting (liquid medium). In this way, their rooting capability were examined. Rooting trials were conducted before stem cuttings were taken to solid rooting media. As conclusions, the highest rooting ratio were obtained for 10 ppm of NAA (94.43%) and 100 ppm of IBA (93.9%) in liquid media. Moreover, the highest root length and the average root length were quite low in liquid media.

KEY WORDS: Fitohormones, vegetative propagation, auxins, rooting

INTRODUCTION
UVOD

Millions of plants are vegetatively produced every year to be used in different fields. If the genetic structure of an individual is intended to be conserved, its production is based on cutting. In this way, the genetic structure of rootstock is conserved exactly. Millions of plants are produced by use of this method every year to be used in various fields such as landscaping, forestry, and agriculture.

Since auxin had been begin to use as plant growth regulators, lots of researches have been conducted and lots of methods have been developed and applied on various plant

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species. Although these researches conducted, auxin application-related researches are needed to grow more quality and productive plants (Chhun *et al.,* 2003).

Rooting of cutting varies from species to species. Various pre-treatments are carried out in order to facilitate the rooting of cutting and increase success. Hormone applications are one of the most common pre-treatments. Research evidence suggests that auxins play a central role in the determination of rooting capacity, by enabling the faster production of rooted cutting material which is essential for vegetative propagation (Fogaça and Fett-Neto, 2005). Auxins are known to increase rooting percentage and rooting time together with uniformity of rooting (Hartmann *et al.,* 2002).

In these applications, the cross section of a plant is exposed to the rooting hormones prepared in high concentrations for a short time. Then cuttings are taken to such rooting media as peat, sand, and pearlite, and root formation is waited. The cuttings taken to rooting media cover extensive areas. So, the rooting media cannot be used for other purposes until this application, which is usually carried out in greenhouses, ends.

The rooting period of plants following the hormone application varies from species to species. It may take from a couple of weeks to twelve weeks or longer (Zencirkıran, 2013). The plant's root development cannot be monitored visually in this period. Rooting percentages may be too low after the applications that take many months. That causes big losses of labor and very high costs.

This study aims to conduct production through cuttings by use of a different method. To this end, contrary to classic applications, cuttings were not exposed to short-time high hormone doses, but they were kept in low-concentration hormones permanently. To compare the research results with the results of conventional applications, common rooting methods were also employed.

MATERIALS AND METHODS
MATERIJALI I METODE

In this study, species *Ficus benjamina* L. cuttings, which were in a height of 8 to 10 cm and prepared from last-year shoots, were used. In the cutting preparation, to keep two buds on each cutting was ensured. The cuttings were kept in pure water to dry and were used in rooting trials in less than 24 hours.

Two different rooting methods were tried in this study. In the first one, the concentrations of IAA, IBA, GA_3, and NAA hormones were prepared in doses of 100, 50 and 10 ppm. Cardboard cups having a volume of nearly 150 mm were filled with these hormones up to half. Then 20 of prepared cuttings were put in each of these media. After putting the cuttings, the hormone level in the cup was marked. In case of any decrease of hormone level in the cup as to daily check, such cups were completed with pure water. By this means, it was tried to prevent any change in hormone concentration that might occur as a result of surface evaporation. The concentrations of IAA, IBA, GA_3, and NAA hormones prepared in the doses of 100, 50 and 10 ppm were kept in a +4 °C refrigerator in the study period. The cups and the hormones inside them were changed once every five days. In this stage of the study, 12 applications were carried out (3 doses from each hormone). Also, a control group was used. The rooting of cuttings in pure water was monitored in the control group.

Rooting trials were conducted also in solid rooting media in order to compare the results of the applications focused on in the study with those of classic applications. 1000, 3000 and 5000 ppm concentrations of IAA, IBA, GA_3, and NAA hormones were tried. The cross sections of the prepared cuttings were made to contact with these hormones for 4 to 5 seconds, and the cuttings were planted in rooting media. Sand, peat, and perlite were used as rooting media. The rooting media were irrigated once every three days, thereby keeping the media humid continuously. In this way, the cuttings exposed to 13 applications (3 doses from each one of 4 hormones and one control group application) were taken to 3 different rooting media. Thus, 39 different application results were compared.

The cuttings subjected to 13 different applications in liquid rooting media and 39 different applications in solid rooting media were kept in rooting media for 45 days. At the end of this period, measurements were carried out on cuttings to determine rooting percentage (%) (RP), the number of roots (RN), the biggest root length value (mm) (RB), the average root length value (mm) (RA) and the average root thickness value (mm) (RT).

All data were analyzed using SPSS for Windows. Firstly, a one-way analysis of variance (ANOVA) was performed on the data. Then Duncan's test was set at the 0.05 confidence level to separate treatment means.

RESULTS
REZULTATI

Different doses of hormones may have different effects on traits. Thus, more reliable results may be obtained if evaluation is made by regarding each dose of each hormone as a separate treatment, that is, a separate treatment. The applications were effective in terms of all the characters ($P<0.05$). There was no rooting in 10 ppm doses of IAA and IBA, and there was rooting only in 100 ppm of GA_3 and in 10 ppm of NAA. The highest rooting percentage was in 10 ppm of NAA (94.43%) and in 100 ppm of IBA (93.9%). These va-

Table 1. Effect of different concentration of fitohormones (IAA, IBA, GA3 and NAA) on rooting traits of *Ficus benjamina* L. in liquid rooting media
Tablica 1. Utjecaj različitih koncentracija fitohormona (IAA, IBA, GA3 i NAA) na značajke zakorjenjivanja reznica vrste *Ficus benjamina* L. u tekućim medijima

Fitohormone *Fitohormon*	Concentration (ppm) *Koncentracija (ppm)*	Rooting percentage (%) RP *Postotak ukorjenjivanja (%) RP*	Number of roots RN *Broj korijena RN*	Biggest root length value (mm) RB *Najveća vrijednost dužine korijena (mm) RB*	Average root length value (mm) RA *Prosječna vrijednost dužine korijena (mm) RA*	Average root thickness value (mm) RT *Prosječna vrijednost debljine korijena (mm) RT*
IAA *IAA*	100	71,43d	10,8d	22,86b	16,686b	0,866b
	50	14,29a	7c	13,02ab	9,505ab	0,890b
IBA *IBA*	100	93,9e	1,57a	43,25c	31,571c	0,934b
	50	57,14c	5,75b	25,41b	18,549b	0,610a
GA_3 *GA_3*	100	57,14c	1,75a	48,00c	35,038c	0,788b
NAA *NAA*	10	94,43e	5,57bc	10,22ab	7,460ab	0,636a
Control *Kontrola*	0	37,56b	3,33ab	4,83a	3,523a	0,493a
F Value *Vrijednost F*		300,949***	14,049***	14,178***	14,178***	13,914***

Means followed by the same letter in a column are not significantly different at P<0.05, based on Duncan's Test.
*** The mean difference is significant at the 0.01 level.

Prosječne vrijednosti s istim slovom u koloni nisu signifikantne pri P<0.05, na temelju Duncan's Testa.
**** Značajno kod 0.01.*

lues were almost threefold of the rooting percentage obtained in the control group (37.56%). When other characters were examined, the control group was in the first homogeneous group in terms of all traits. That indicates that hormone applications have positive effects on all traits. The highest values were obtained in 100 ppm IBA, in terms of RB and RT; in 100 ppm GA_3 and 100 ppm IBA in terms of RB; and in 100 ppm IAA in terms of RN (Table 1).

To compare the obtained values with classic rooting trials, the rooting values of the cuttings in the solid rooting media were examined. There were significant differences in RP, RN, RB, and RT of species *Ficus benjamina* L. cuttings (P<0.05). The highest RP (70.51%) was obtained in the perlite; whereas 53.19% and 39.17% of RP were obtained for peat and sand media, respectively. The highest RB and RT values were obtained in the sand medium (Table 2).

There were significant differences among the five groups (IAA, IBA, GA_3, NAA, and control) in RP, RN, RB, RA, and RT (P<0.05). The highest RP were obtained in 5000 ppm IAA (80.15%) and 3000 ppm IAA (77.21%) applications. The highest two values in terms of the number of roots were obtained in 3000 ppm NAA (9 pcs) and 5000 ppm NAA (7.7 pcs) applications. While the biggest root length values were obtained in 3000 ppm IBA (51.77 mm), 3000 ppm IAA

Table 2. Rooting traits of *Ficus benjamina* L. in three different solid rooting media
Tablica 2. Značajke zakorjenjivanja vrste *Ficus benjamina* L. u tri različita kruta supstrata

Media *Medij*	Rooting percentage (%) RP Postotak ukorjenjivanja (%) RP	Number of roots RN *Broj korijena RN*	Biggest root length value (mm) RB *Najveća vrijednost dužine korijena (mm) RB*	Average root length value (mm) RA *Prosječna vrijednost dužine korijena (mm) RA*	Average root thickness value (mm) RT *Prosječna vrijednost debljine korijena (mm) RT*
Sand *Pijesak*	39,167a	5,43b	54,2836b	29,1821a	,885b
Peat *Treset*	53,194b	2,85a	34,7875a	24,8208a	,860b
Pearlite *Perlit*	70,506c	5,27b	40,1427a	26,0459a	,679a
F Value *Vrijednost F*	41,680***	11,257***	11,884***	1,390	18,627***

Means followed by the same letter in a column are not significantly different at P<0.05, based on Duncan's Test.
*** Značajno kod 0.01 level.

Prosječne vrijednosti s istim slovom u koloni nisu signifikantne pri P<0.05, na temelju Duncan's Testa.
**** Značajno kod 0.01.*

Table 3. Effect of different concentration of Auxins (IAA, IBA, GA3 and NAA) on rooting traits of *Ficus benjamina* L. in different solid substrates
Tablica 3. Utjecaj različitih koncentracija auksina (IAA, IBA, GA3 i NAA) na značajke zakorjenjivanja vrste *Ficus benjamina* L. u različitim krutim supstratima

Fitohormone Fitohormon	Concentration (ppm) Koncentracija (ppm)	Rooting percentage (%) RP Postotak ukorjenjivanja (%) RP	Number of roots RN Broj korijena RN	Biggest root length value (mm) RB Najveća vrijednost dužine korijena (mm) RB	Average root length value (mm) RA Prosječna vrijednost dužine korijena (mm) RA	Average root thickness value (mm) RT Prosječna vrijednost debljine korijena (mm) RT
IAA IAA	5000	80,15e	3,0ab	50,01d	31,5e	0,71bc
	3000	77,21e	6,5cde	50,05d	30,1de	0,79bc
	1000	41,67bc	2,0a	35,71bcd	27,2cde	0,59ab
IBA IBA	5000	39,29bc	4,8abcd	38,29bcd	21,0bcde	0,70bc
	3000	68,75de	5,6bcd	51,77d	32,3e	0,78bc
	1000	68,06de	2,1a	23,36ab	20,3bcde	0,73bc
GA_3 GA_3	5000	12,5a	4,0abc	22,04ab	16,1abc	0,61ab
	3000	42,5bc	2,8ab	15,36a	12,5ab	0,44a
	1000	37,5bc	2,0a	15,52a	4,8a	0,54ab
NAA NAA	5000	52,08cd	7,7de	41,76bcd	28,1cde	1,04d
	3000	25ab	9,0e	44,67cd	22,6bcde	0,87cd
	1000	46,88c	3,0ab	28,73abc	18,2bcd	0,67abc
Control *Kontrola*	0	49,9c	2,2ab	22,42ab	14,0ab	0,64abc
F Value *Vrijednost F*		$19{,}155^{***}$	$9{,}369^{***}$	$8{,}363^{***}$	$6{,}040^{***}$	$7{,}792^{***}$

Means followed by the same letter in a column are not significantly different at P<0.05, based on Duncan's Test.
*** The mean difference is significant at the 0.01 level.

Prosječne vrijednosti s istim slovom u koloni nisu signifikantne pri P<0.05, na temelju Duncan's Testa.
*** *Značajno kod0.01.*

(50.05 mm), and 5000 ppm IBA (50.01) applications, the biggest RA values were obtained in 3000 ppm IBA (32.3 mm), 5000 ppm IAA (31.5 mm), and 5000 ppm IBA (30.1) applications. The highest RT value was obtained in 5000 ppm NAA application (Table 3).

The results of low-concentration hormone applications in solid and liquid rooting media are shown on Table 4. Regarding, rooting percentage, the number of roots, and root thickness, it is clear that these values were quite close to one another, but the biggest root height value and the average root height value were quite low in liquid rooting media. Although there were no significant differences between rooting media in RN and RT, there were significant differences in RP, RB, and RA (Table 4). The biggest root height value and the average root height value were found to be higher in solid rooting media, while rooting percentage was found to be higher in liquid rooting media.

It is seen that liquid rooting media are more advantageous than solid rooting media in terms of many characters (Table 4). The highest rooting percentage in solid rooting media was 80.15% (5000 ppm IBA (Table 3), while the highest rooting percentage in liquid rooting media was over 90% (94.43% for 10 ppm NAA; 93.9% for 100 ppm IBA) (Table 1). While the biggest number of roots was 9 in solid rooting media (Table 3), it was up to 10.8 in liquid rooting media (Table 1). Similar results were obtained in terms of other characters, too. The biggest root height value was found to be 50.05 mm for 3000 ppm IAA in solid rooting media (Table 3) and was found to be 48 mm for 100 ppm GA_3 in liquid rooting media (Table 1). While the highest RB value was found to be 31.5 mm in 5000 ppm IAA in solid rooting media (Table 3), it was found to be 35.04 mm in 100 ppm GA_3 in liquid rooting media (Table 1). The biggest root thickness value was found to be 1.04 mm in solid rooting media (Table 3), but it was 0.934 mm in liquid rooting media (Table 1).

DISCUSSION
RASPRAVA

Various studies on the propagation of *Ficus* species via cutting have examined the effects of different cutting extracting periods, rooting media, hormone applications, etc. on the rooting of cuttings. Küden *et al.* (1993) found out that cutting extracting periods, rooting media, and IBA applications affected rooting rates in cuttings, and the rooting percentages varied between 0-90% in their applications. Tekintaş and Seferoglu (1998) conducted rooting trials on *Ficus carica* in different media and obtained the highest rooting rate in the sand medium (71%). It was followed by peat (31%), pearlite (27%), and soil (25%) respectively. Antunes

Table 4. One-way analysis of variance (ANOVA) for effects of solid and liquid rooting media on rooting traits of *Ficus benjamina* L.
Tablica 4. Jednostruka analiza varijance (ANOVA) za utjecaj krutih i tekućih medija za zakorjenjivanje na značajke zakorjenjivanja vrste *Ficus benjamina* L.

Substrat *Supstrat*	Rooting percentage (%) RP *Postotak ukorjenjivanja (%) RP*	Number of roots RN Broj korijena RN	Biggest root length value (mm) RB *Najveća vrijednost dužine korijena (mm) RB*	Average root length value (mm) RA *Prosječna vrijednost dužine korijena (mm) RA*	Average root thickness value (mm) RT *Prosječna vrijednost debljine korijena (mm) RT*
Solid *Kruti*	60,68	4,72	41,23	26,28	0,76
Liquid *Tekući*	61,62	4,87	26,12	19,07	0,75
F Value *Vrijednost F*	5,763*	0,106	33,853***	16,490***	0,037

*The mean difference is significant at the 0.05 level.
***The mean difference is significant at the 0.01 level.

* *Značajno kod 0.05*
*** *Značajno kod 0.01*

et al. (1996) examined the effects of different stratification periods, IBA concentrations, and rooting media on the rooting of *Ficus carica* cuttings and determined that the best root and shoot development was obtained in the cuttings subjected to a dose of 100 ppm of the IBA hormone and planted in sand: soil mixture in the ratio of 1:1 without any stratification. A study was conducted on the propagation of *Ficus carica* via green cutting. Green cuttings with 2 to 3 leaves exposed to a dose of 1000 to 4000 ppm of the IBA hormone were planted in stream sand, and a rooting of 85% to 100% was obtained (Kai *et al.,* 1997). In addition, some studies on the propagation of *Ficus carica* via tissue culture conducted in recent years have yielded favorable results (Demiralay *et al.,* 1998; Günver and Ertan; 1998; Kumar *et al.,* 1998; Nobre and Romano, 1998). However, all of these studies have required more time, labor, materials, and hormones in comparison to the method employed in the present study and do not have success rates higher than the one obtained in the present study.

Many studies have focused on the effects of auxin group hormones on rooting and plant development. Alvarez *et al.* (1989) examined the effects of IAA and IBA in *Malus pumila*; Şevik and Güney (2013a, 2013b) examined the effects of IAA, IBA, NAA and GA_3 in *Melissa officinalis*; Stefancic *et al.* (2005) examined the effects of IAA and IBA in *Prunus* spp., and Chhun *et al.* (2003) examined the effects of IAA, IBA, and NAA in *Oryza sativa*. The previous studies mostly show that auxin group hormones are influential on rooting. That is consistent with the results of the present study.

Gibberellins are the third most commonly used plant hormones with a share of 17%. The most commonly used commercial gibberellin is GA_3. It is mostly used for increasing the height of a plant or flower yield (Kumlay and Eryiğit, 2011). The present study demonstrated that rooting percentage, root height, and root thickness values were 1.5 to 9.9 times higher among cuttings exposed to a dose of 100 ppm of the GA_3 hormone in comparison to the control group (Table 1). That is consistent with the results provided in the literature, too (Sevik and Guney, 2013a, 2013b).

CONCLUSIONS
ZAKLJUČCI

Liquid rooting media provide bigger advantages in comparison to the conventional applications. They allow monitoring the course of rooting of plants and prevent occupying solid rooting media in vain because plants whose roots have grown enough are taken to solid rooting media. Liquid rooting media also allow producing many plants in a limited rooting area. For example, in the present study, cuttings were placed in solid rooting media at the intervals of 2 cm, and an area of approximately 400 cm^2 was used for 100 cuttings. On the other hand, 20 cuttings were placed in each cardboard cup which had a diameter of almost 6.5 cm in liquid rooting media. An area of nearly 43 cm^2 was used for 100 cuttings. Accordingly, liquid rooting media allow the rooting of the same amount of cuttings as solid rooting media in an area of almost 1/10 of the area used by solid rooting media. Another advantage of the employed method is fewness of the number of materials used. While classic methods require flowerpots and rooting platforms covering wide areas as well as such materials as sand, pearlite, or peat as a rooting medium, the method employed in the present study uses only cardboard cups and pure water and requires smaller amount of hormones. Since the individuals whose root formation has reached the adequate level in liquid rooting media are placed in flowerpots, success rate is close to 100%. Therefore, this method, which is easier and cheaper, can be effectively used in the fields where many individuals such as medical plants, aromatic plants, ornamental plants, and field crops need to be rooted. However, further studies should be carried out to determine the hormone and the concentration that yield the best result for each plant.

According to the results of the present study, different hormones and different concentrations have different effects on rooting percentage and morphological characters. Thus, hormones should be selected based on the primary character. For instance, 10 ppm NAA should be preferred when high rooting rate is requested; 100 ppm IAA should be preferred when a big amount of root formation is requested; and 100 ppm GA_3 should be preferred when long root is requested.

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Sažetak

Tehnike vegetativne proizvodnje od kojih se metoda reznica uvelike koristi u razmnožavanju ukrasnog bilja, imaju ključnu ulogu za očuvanje genetskih izvora biljaka. Mjesto uzimanja reznice na biljci, medij ukorjenjivanja i fitohormon ukorjenjivanja neki su od ključnih čimbenika koji utječu na uspjeh razvoja zakorjenjivanja reznica od stabljike. Primarni cilj ovog istraživanja bio je utvrditi najbolje koncentracije fitohormona i idealni medij zakorjenjivanja za reznice od stabljike vrste *Ficus benjamina* L. Konvencionalno, najčešća metoda vegetativnog razmnožavanja je zakorjenjivanja pupova u različitim supstratima kao što su perlit, treset, pijesak do izloženosti visoko koncentriranim hormonima zakorjenjivanja (IBA, IAA, NAA, itd.). Ali ta konvencionalna tehnika zahtijeva široka područja u fazi zakorjenjivanja masovne produkcije, sprječava nadzor tijeka zakorjenjivanja i traži visoku količinu materijala koji se koriste kao hormoni i mediji zakorjenjivanja. U ovom istraživanju, ispitana je nova metoda koja bi mogla biti poželjnijom u masovnoj proizvodnji biljaka. Izvršeno je 39 različitih tretiranja i prikazani su njihovi rezultati. Pijesak i perlit korišteni su kao kruti medij zakorjenjivanja. Pupovi su se trajno čuvali u hormonima niske koncentracije nakon rezanja (tekući medij). Na taj način je ispitana njihova sposobnost zakorjenjivanja. Ispitivanja zakorjenjivanja provedena su prije nego su pupovi odnijeti u kruti medij zakorjenjivanja. Kao zaključak, dobiveni su najviši omjeri zakorjenjivanja za 10 ppm NAA (94.43%) te 100 ppm IBA (93.9%) u tekućim medijima. Najveća duljina korijena i prosječna duljina korijena bile su prilično male u tekućim medijima.

KLJUČNE RIJEČI: Fitohormoni, autovegetativno razmnožavanje, auksini, zakorjenjivanje