The variations of physiological and antioxidant properties in *Schefflera arboricola* cuttings over the rooting period

Maryam FATHI
Shahram SEDAGHATHOOR (✉)
Seydeh Khadije ABBASNIA

Summary

Schefflera arboricola (Hayata) Merr. is propagated by leafy stem cuttings. In order to study the biochemical variations and vegetative traits of *S. arboricola* cuttings over the rooting period, an experiment was carried out on the basis of a randomized complete block design with 13 treatments and three replications. The experimental treatments were composed of indole-3-butyric acid (IBA), naphthaleneacetic acid (NAA), abscisic acid (ABA), salicylic acid (SA) and their combinations at different rates. It was found that hormone application had a favorable impact on rooting and increased rooting percentage in most treatments when compared to control. The application of different hormones influenced morphological traits to varying degrees. According to the findings, the highest rooting percentage was obtained from cuttings treated with 1000 mg L^{-1} NAA, the highest number of roots was obtained from those treated with 1000 mg L^{-1} NAA + 200 mL L^{-1} SA, and the longest roots from those treated with 1000 mg L^{-1} NAA or 100 µM ABA. Significant differences were observed between two studied ABA rates in chlorophyll contents and peroxidase content. The highest catalase activity was observed in cuttings treated with 1000 mg L^{-1} IBA + 50 µM ABA + 200 mL L^{-1} SA.

Key words

abscisic acid, salicylic acid, naphthaleneacetic acid, umbrella plant

Department of Horticulture, Rasht branch, Islamic Azad University, Rasht, Iran

✉ Corresponding author: sedaghathoor@yahoo.com

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Introduction

Schefflera arboricola is a member of the family Araliaceae (Mazhar et al., 2011). Besides stem cuttings it can be propagated also by leafy stem cuttings (Şevik et al., 2015) where plant hormones, especially auxin, play an important role by affecting the rooting rate and percentage of the rooted cuttings. Plants produce natural auxin in their young branches and leaves, but viable rooting requires the application of artificial auxin (Kasim and Rayya, 2009). The cuttings of some plant species bear roots readily because the indigenous auxin produced in their tips is mobilized to the end of cuttings. The application of exogenous auxin to these cuttings can increase the number of roots remarkably (Nordstrom et al., 1991). Salicylic acid (SA) is a strong plant hormone whose exogenous application to plants can influence the growth and development, crop production, and resistance to diseases and environmental stresses like salinity, drought, UV irradiation, and heavy metals (Wilson and van Staden, 1990).

Abscisic acid (ABA) plays an essential role in plant responses to environmental stresses (Seo and Koshiba, 2002). It has been shown that hormone overuse during rooting can disrupt the hormone balance in plants in addition to its costs. Therefore, there is no question about the significance of finding the best hormone rate for the propagation of ornamental species (Ersoy and Aydin, 2008).

In a study on *S. arboricola*, the cuttings were treated with 1000, 3000, or 5000 ppm Indole acetic acid (IAA), indole-3-butyric acid (IBA), naphthalene acetic acid (NAA), or gibberellic acid (GA). The highest rooting percentage (75 percent) was reported for cuttings treated with 5000 ppm NAA, and the highest root diameter was obtained from those treated with 5000 ppm NAA. The treatment with 1000 ppm gibberellic acid was related to the highest root number and average root length and the treatment with 1000 ppm IBA yielded the highest root length (Şevik et al., 2015). A study showed that the application of IBA influenced the rooting potential of a species *S. arboricola*, *Ficus benjamina* and *Syringa amurensis* (Karimian et al., 2013). A study on *Cordyline terminalis* revealed that the highest root dry weight, root fresh weight, root length, and branch number were obtained from the plants treated with a mixture of 2000 mg L^{-1} NAA and 1000 mg L^{-1} IBA (Rahdari et al., 2014).

Total phenol content was higher in cuttings treated with IBA, especially at the initiation and expansion stages (Rout, 2006). It has been reported that SA can improve the rooting of cuttings in some woody and herbaceous species under *in vivo* conditions, especially when it is accompanied with auxin (Bojarczuk and Jankiewicz, 1975; Kling and Meyer, 1983). But, under *in vitro* conditions, SA restrains the rooting induced by auxin in 1 mm pieces of apple branch via increasing the oxidation of auxin during auxin-sensitive phase (24 - 96 hours) (De Klerk, 1996). ABA controls root elongation by adjusting cell length. In corn plants exposed to water stress, ABA stimulates root elongation by increasing the length of elongating region and reducing the accumulation of reactive oxygen species (Sharp et al., 2004). Given the importance of this subject, in the present study different rates of IBA, NAA, ABA, and SA were applied to explore the biochemical changes during rooting and to identify their optimum rate for commercial propagation of *S. arboricola*.

Materials and Methods

Plant material and experimental design

The present study aimed to explore the variations of antioxidant enzymes, chemicals and vegetative traits of *Schefflera arboricola* cuttings during rooting period. The cuttings were obtained from three years old stock plants that were grown in Rostami greenhouse, Rasht, Iran. The experiment was based on a randomized complete block design with 13 treatments (T_1 = control; T_2 = 1000 mg L^{-1} IBA; T_3 = 2000 mg L^{-1} IBA; T_4 = 1000 mg L^{-1} NAA; T_5 = 2000 mg L^{-1} NAA; T_6 = 50 µM ABA; T_7 = 100 µM ABA; T_8 = 200 mL L^{-1} SA; T_9 = 300 mL L^{-1} SA; T_{10} = T_2 + T_6; T_{11} = T_8 + T_{10}; T_{12} = T_4 + T_8; T_{13} = T_6 + T_{12}; Table 1) in three replications. The study was carried out in a greenhouse in Rasht County (Kishestan) in Guilan Province, Iran. *S. arboricola* cuttings were smeared with the hormones and were planted in a mixture of sand and perlite (1:1). The bases of the cuttings were kept in the pre-prepared treatments for 10 seconds. Each experimental plot was composed of two pots, each one containing one cutting. The mean temperature was 22 - 25°C and mean relative humidity was 70 - 80%. The plots were irrigated by fog system. The observations of the response to rooting were recorded 75 days after hormones application.

Table 1. The experimental treatments studied on *Schefflera arboricola* cuttings

Code	Description
T_1	Control
T_2	1000 mg L^{-1} IBA
T_3	2000 mg L^{-1} IBA
T_4	1000 mg L^{-1} NAA
T_5	2000 mg L-1NAA
T_6	50 µM ABA
T_7	100 µM ABA
T_8	200 mL L^{-1} SA
T_9	300 mL L^{-1} SA
T_{10}	1000 mg L^{-1} IBA + 50 µM ABA (T_2 + T_6)
T_{11}	1000 mg L^{-1} IBA + 50 µM ABA + 200 mL L^{-1} SA (T_{10} + T_8)
T_{12}	1000 mg L^{-1} NAA + 200 mL L^{-1} SA (T_4 + T_8)
T_{13}	1000 mg L^{-1} NAA + 200 mL L^{-1} SA + 50 µM ABA (T_{12} + T_6)

Recorded traits

The experimental treatments were taken out of the substrate simultaneously and the rooted cuttings were counted to determine the rooted cuttings percentage as one of the main criteria of treatment assessment. To find out fresh weight, the roots were detached from the cuttings and were weighed with a 0.001-g-precision scale. Then, they were separately oven-dried at 95°C for 24 hours to estimate the dry weight. The rooting percentage was calculated as the ratio of the number of rooted, healthy cuttings to the total number of cuttings. The number of roots was determined by their visual counting. Root area (cm) was estimated by the Atkinson (1991)'s method as below:

$$\text{Root area} = (\text{root length} \times 3.14 \times \text{root volume} \times 2)^{0.5}$$

in which, to determine root volume, they were detached from the cuttings and then, they were placed in a graduated cylinder containing water. The difference in volume before and after the roots were put in the cylinder represented their volume. Root length is a good measure of its growth and development because of its contact with soil. So, we recorded the root length as well as the maximum and minimum root length for each cutting.

Chlorophyll content

Total chlorophyll and chlorophyll *a* and *b* contents of leaves were measured with a spectrophotometer. First, 0.5 g of leaves was ground in a mortar. Then, 100 mL of 80% acetone was added in three steps, and each time, the solution was ground, infiltrated, and read with a spectrophotometer (Mazumdar and Majumder, 2003).

$$\text{Chlorophyll } a = (9.93 \times A_{660} - 0.777 \times A_{643})V.100W$$

$$\text{Chlorophyll } b = (17.6 \times A_{643} - 2.81 \times A_{660})V.100W$$

$$\text{Total Chlorophyll (mg/g)} = (7.12 \times A_{660} + 16.8 \times A_{643})V.100W$$

in which:

V = the volume of infiltrated solution (derived from centrifuge),

A = light absorption at 663 and 645 nm,

W = sample fresh weight (g).

Antioxidant property

The antioxidant capacity of the leaves was derived from the free radical scavenging property of DPPH (diphenyl picryl hydrazyl). In this procedure, 1 g of the sample was ground in a Chinese mortar containing 100 mL of 85% methanol. Then, it was placed at room temperature for 1 h. After that, the solution was infiltrated and centrifuged at 3000 rpm for 5 min. Finally, 0.5 mL of the surface phase was taken with a sampler, 2.5 µL of DPPH was added to it, and the solution was placed in darkness for 20 min. Then, its absorption was read at 517 nm with a spectrophotometer (Ramandeep and Savage, 2005).

The inhibitory of DPPH percentage was calculated as below:

$$\%\text{Inhibitio n} = ((A - A_b) / A_0) \times 100$$

in which:

A = absorption (sample + DPPH),

A_0 = DPPH absorption,

A_b = sample absorption.

The procedure to prepare 50 mM phosphate buffer with the pH of 7 was as follows: 0.68 g of potassium dihydrogen phosphate (KH_2PO_4) salt, as well as 2 g of PVPP and Na-EDTA, was dissolved in 50 mL of distilled water and its volume was adjusted to 100 mL. Then, 0.87 g of potassium monohydrogen phosphate (K_2HPO_4), as well as 2 g of PVPP and Na-EDTA, was dissolved in 50 mL of distilled water and its volume was adjusted to 100 mL. These two solutions were taken as the stock solution. For each assay, 39 mL of the first solution was mixed with 61 mL of the second solution, and its pH was adjusted to 6.8-7.2 with a pH meter.

Peroxidase (POD) activity was measured by the Addy and Goodman (1972) procedure and catalase (CAT) enzyme activity was measured by the Khoo et al. (2011) procedure.

Data collected about morphological traits and chemical properties were analyzed on the basis of a randomized complete block design with three replications using MSTATC software package (Freed et al., 1989). Also, means were compared by the LSD test at $p < 0.05$.

Results and discussion

Hormonal treatments affected rooting properties

Analysis of variance showed that the experimental treatments influenced all traits significantly at $p<0.05$ or $p<0.01$ level (Table 2). Control cuttings exhibited the lowest rooting percentage of 41.67% (Figure 1). Two IBA rates showed insignificant differences in rooting percentage (Figure 1). This does not agree with the results of Karimian et al. (2013) and Nautiyal et al. (2015) who found that IBA can be applied in any rate to facilitate the rooting of the cuttings, but higher rates give better results. Nautiyal et al. (2015) studied the effect of IBA at two rates of 2000 and 4000 ppm on the rooting of *S. arboricola*. They reported that rooting was 65% in plants treated with 4000 ppm IBA, 40% in those treated with 2000 ppm IBA, and at least 20% in control (untreated plants).

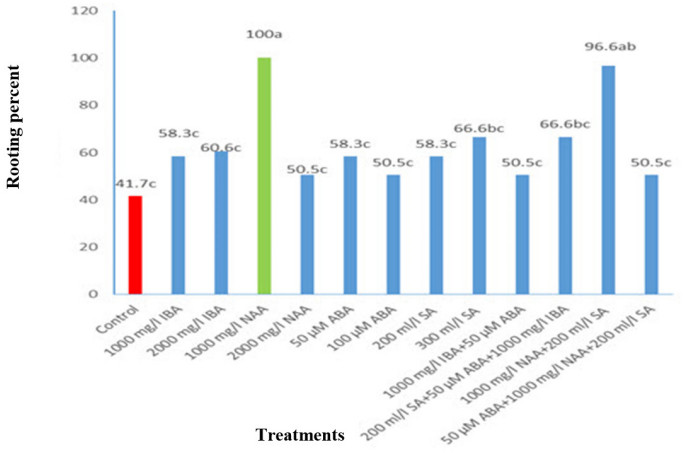

Figure 1. The effect of experimental treatments on the rooting percentage of *Schefflera arboricola* cuttings

Two studied rates of NAA exhibited significant differences in rooting percentage, antioxidants content, root length, the number of root branches, root area, root dry weight, and root fresh weight (Table 2). The highest root fresh and dry weights were obtained from the plants treated with 1000 mg L^{-1} NAA among all studied treatments. Similarly, Abu-Zahra et al. (2011) reported for *S. arboricola* that all studied auxin rates improved rooting as compared to control; the best results for rooting were observed in leaf bud cuttings treated with 2000 ppm NAA. It should be noted that in our study, cuttings treated with 2000 mg L^{-1} NAA produced the least number of root branches. Şevik et al. (2015) reported the highest root diameter and rooting percentage (75%) in the treatment of 5000 ppm NAA.

The SA containing treatments with T_{10} and T_{11} were related to the lowest root fresh and dry weights (Table 3).

Table 2. Analysis of variance of the studied traits as influenced by the experimental treatments

S.O.V.	df	Rooting percentage	Longest root	Shortest root	Branches (Shoots)	Root area	Dry weight	Fresh weight	Chlorophyll *a*	Chlorophyll *b*	Total chlorophyll	Catalase	Peroxidase	Antioxidant
Replication	2	110.26^{ns}	1329.56^{ns}	61.14*	39.95*	146.01^{ns}	0.56*	5.33^{ns}	0.54^{ns}	0.083^{ns}	0.82^{ns}	0.051^{ns}	0.12^{ns}	33531.18*
Treatment	12	930.77**	355436*	46.26**	63.37*	693.46*	0.49*	11.99**	6.67**	1.65**	14.21**	0.29**	0.34**	141914.3*
Error	24	331.09	1355.41	12.92	14.36	213.89	0.20	3.944	2.048	0.47	4.14	0.034	0.08	9490.96
CV (%)		29.32	51.73	29.69	61.78	55.79	89.27	64.26	25.74	23.5	24.24	19.14	23.97	40.85

** - significant at $p < 0.01$; * - significant at $p < 0.05$; ns - non-significant

Table 3. Means comparison of the studied traits as influenced by the experimental treatments

Treatments	Longest root (mm)	Shortest root (mm)	Branches (Shoots)	Root area (mm^2)	Dry weight (g)	Fresh weight (g)	Chlorophyll *a* (mg/FW)	Chlorophyll *b* (mg/FW)	Total chlorophyll (mg/FW)	Catalase (nM/g FW)	Peroxidase (nM/g FW)	Antioxidant (µg/mL)
T_1, control	70.94 abcd	9.00 cde	3.33 cde	22.38 cd	0.49 bc	3.38bcd	8.4a	4.10 a	12.5 a	0.71 def	1.16 abcd	158 b
T_2, 1000 mg/L IBA	77.18 abc	10.70 bcd	9.67 abc	25.11 bcd	0.36 bc	2.55 cd	6.73 ab	3.13 abc	9.22 abc	0.75 def	1.24 abc	218 b
T_3, 2000 mg/L IBA	97.42 ab	16.56 ab	4 cde	14.27 d	0.25 c	]1.91 cd	5.84 bc	3.05 abc	8.89 bcde	0.98 cd	1.56 ab	147 b
T_4, 1000 mg/L NAA	123.1 a	12.06 abcd	7.67 cde	48.42 ab	1.05 ab	7.29 a	6.13 ab	3.34 ab	9.47 abc	0.64 ef	1.66 a	949.4 a
T_5, 2000 mg/L NAA	27.73 cd	12.32 abcd	1 e	18.33 cd	0.181 c	1.73 cd	6.27 ab	3.23 ab	9.5 abc	0.68 cde	1.53 ab	166.3 b
T_6, 50 µM ABA	67.97 a-d	9.723 cde	13.67 ab	16.55 d	1.05 ab	5.92 ab	6.99 ab	4.17 a	11.17 ab	0.92 cde	1.09 bcd	201.7 b
T_7, 100 µM ABA	102.2 a	13.80 abc	6.66 cde	20.46 cd	0.56 abc	3.33 bcd	3.33 d	2.33 bcd	5.67 def	0.97 cd	1.61 a	151 b
T_8, 200 mL/L SA	72.45 a-d	17.12 a	3.33 cde	24.28 bcd	0.34 bc	1.93 cd	5.43 bcd	2.67 bcd	7.77 b-f	1.31 b	0.75 cd	239 b
T_9, 300 mL/L SA	39.69 bcd	12.56 a-d	3.67 cde	15.80 d	0.11 c	1.01 d	4.74 bcd	2.35 bcd	7.10 cdef	1.20 bc	1 cd	153.3 b
T_{10}, T_2+T_6	10.50 d	4.333 e	0.50 e	13.87 d	0.051 c	1.14 d	3.5 cd	2 cd	5.5 ef	0.56 f	1.23 abc	184.7 b
T_{11}, T_8+T_{10}	33.26 cd	7.09 de	2.67 de	16.58 d	0.14 c	0.89 d	5.48 bcd	2.91 bc	8.39 b-f	1.71 a	0.70 d	250 b
T_{12}, T_4+T_8	102.2 a	14.84 abc	14.67 a	42.22 abc	1.29 a	4.88 abc	5.89 bc	3.09 abc	8.97 bcd	1.13 bc	0.83 cd	183 b
T_{13}, T_6+T_{12}	100.4 ab	17.27 a	9.67 abc	62.54 a	0.6883 abc	4.19 abcd	3.49 cd	1.62 d	5.11 f	0.96 cd	1.55 ab	99 b

The means followed with the same letter(s) do not differ significantly

Plants treated with 50 µM ABA + 1000 mg L^{-1} NAA + 200 mL L^{-1} SA (T_{13}) showed the highest root area (Table 3). El-Shrairy and Hegazi (2009) stated that the application of acetylsalicylic acid and indole-3-butyric acid improved the growth of peas which was significantly correlated with the increase in leaf chlorophyll content.

Chlorophyll contents and antioxidant enzymes activity influenced by hormones

Significant differences were observed between two studied ABA rates in chlorophyll *a* and *b*, total chlorophyll contents, and peroxidase content. Cuttings treated with 100 µM ABA had the highest root length, the highest peroxidase activity, and the lowest chlorophyll *a* content. The highest chlorophyll *b* content was related to the treatment of 50 µ ABA. In total, treatment with ABA enhanced root fresh and dry weights. The response of root growth of the plants to environmental variations is governed by developmental genetic programs. ABA hormone is an intermediate for the response to various environmental factors like the presence of nitrate in soil, water stress, and salinity stress. Root endodermis is the site where ABA controls root elongation. Although ABA signaling happens in all cellular layers of the root, research shows that ABA signaling in root endodermis is critical for the regulation of root growth (Haris, 2015).

SA is a mono-phenol that boosts IAA oxidation (De Klerk et al., 1999). Two applied SA rates did not show any significant differences in any measured trait (Table 3). Cuttings treated with 200 mL L^{-1} SA had the shortest roots (T_8 and T_{13}), high root dry weight (T_{12}), proper long root length (T_{12}), and low antioxidant content (T_{13}). The cuttings treated with 300 mL L^{-1} SA (T_9) exhibited low root area and low root fresh and dry weights (Table 3).

Among the treatment combinations, the simultaneous application of 1000 mg L^{-1} IBA and 50 µM ABA (T_{10}) gave the lowest levels of the measured traits. The simultaneous application of 1000 mg L^{-1} IBA, 50 µM ABA and 200 mL L^{-1} SA resulted in the highest chlorophyll *a* content as well as low root fresh and dry weights and root area. The simultaneous application of 1000 mg L^{-1} NAA and 200 mL L^{-1} SA had the longest roots, the highest number of branches, and the highest root dry weight. Finally, the simultaneous application of 1000 mg L^{-1} NAA, 50 µM ABA, and 200 mL L^{-1} SA exhibited the lowest chlorophyll *b* and total chlorophyll contents, the highest short root length, and the highest root area.

The longest roots (123.1 mm) were produced in cuttings treated with 1000 mg L^{-1} NAA (Table 3) followed by treatments T_7, T_{12}, T_{13}, T_3, and T_2. These treatments did not differ significantly as shown in Table 3. The shortest roots (10.5 mm) were related to the treatment with 1000 mg L^{-1} IBA + 50 µM ABA so that this treatment produced the finest roots (Table 3). Cuttings treated with 1000 mg L^{-1} IBA exhibited the highest mean root fresh weight of 7.29 g (Table 3) which was significantly higher than that of control. The lowest mean fresh weight of 0.89 g was observed in cuttings treated with 1000 mg L^{-1} IBA + 50 µM ABA + 200 mL L^{-1} SA (T_{11}) (Table 3). The treatment of cuttings with 1000 mg L^{-1} NAA + 200 mL L^{-1} SA (T_{12}) was related to the highest average root dry weight of 1.29 g. This was significantly higher than that of control (Table 3).

It was found that chlorophyll *a* and *b* and total chlorophyll contents were the highest in control and 50 µM ABA (Table 3). The lowest chlorophyll *a* content was observed in the treatment with 100 µM ABA and the lowest chlorophyll *b* content was related to the treatment of 1000 mg L^{-1} NAA + 50 µM ABA + 200 mL L^{-1} SA (T_{13}). Chlorophyll *b* content was the highest in the treatment of 50 µM ABA as well as in control. Overall, total chlorophyll content was the highest in control and in the treatment with 1000 mg L^{-1} NAA + 50 µM ABA + 200 mL L^{-1} SA (T_{13}). These results are supported by some previous studies. In Ozfidan et al. (2013), the treatment with only 50 µM ABA had no remarkable effect on total chlorophyll content in *Arabidopsis*. Treatment with NAA reduced leaf chlorophyll in *Momordica charantia* slightly and GA3 increased it to some extent, but these fluctuations were not considerable (Tolentino and Cadiz, 2005). It has been shown that when compared to control, the application of NAA at rates ranging 0.25-4 mg L^{-1} caused diverse effects on growth, chlorophyll content, and protein in *Chlorella vulgaris*, but among all NAA rates, 2 mg L^{-1} had the strongest impact (Lv et al., 2006).

Antioxidant enzymes shape the immune system of the plants against biotic and abiotic stresses (Abdelkadar and Ahmed, 2012). The highest catalase enzyme activity was observed in cuttings treated with 1000 mg L^{-1} IBA + 50 µM ABA + 200 mL L^{-1} SA (T_{11}). The lowest catalase enzyme activity was displayed by cuttings treated with 1000 mg L^{-1} IBA + 50 µM ABA (T_{10}) (Table 3).

The highest peroxidase enzyme activity was in cuttings treated with 1000 mg L^{-1} NAA and then, in those treated with 100 µM ABA. The next ranks of peroxidase activity were related to the treatments of 2000 mg L^{-1} IBA, 1000 mg L^{-1} NAA, 50 µM ABA, 2000 mL L^{-1} SA, and 2000 mg L^{-1} NAA. Seemingly, NAA presence plays a significant role in peroxidase enzyme activity. The lowest peroxidase activity was in the treatment with 1000 mg L^{-1} IBA + 50 µM ABA + 200 mL L^{-1} SA (T_{11}), whilst this treatment exhibited the highest catalase activity (Table 3).

In our study, 100 µM ABA exhibited the highest peroxidase enzyme activity and 50 µM ABA resulted in its moderate activity. Peroxidase activity in barley stem was at a plateau during early growth stage without the treatment of ABA, but ABA treatment enhanced its activity. In addition, peroxidase activity was increased at higher rates of ABA and its analogs (Basra et al., 1992).

The loss of peroxidase activity in the presence of NAA can be attributed to its inhibitory effect on its biosynthesis pathway. NAA can interfere at protein synthesis level. The loss of chlorophyll content in the presence of NAA may imply that NAA might have an inhibitor effect on the biosynthesis of tetrapyrrole-containing compounds, resulting in reduced peroxidase activities (Boeuf et al., 2001). Our findings about NAA treatments are inconsistent with other research. This may relate to the differences in plant species, treatment application, and/or treatment duration.

In the present study, hormone application had a desirable impact on rooting percentage, but different levels of the hormones had various effects on different traits. These results can be useful for commercial production of *S. arboricola*. We used 10 cm cuttings with two terminal leaflets and all other leaflets detached. However, a high rooting percentage was obtained. This implies that we can propagate a lot of plants from one single plant if appropriate culture medium is provided and certain dosages of

hormones are applied. Şevik et al. (2015) used one leaf along with its stalk as the cutting. The advantage of this method is that the stem is not marred, and plant shape is preserved. Furthermore, by this method, the number of cuttings we can take from one single plant is not limited.

The results revealed that the hormones influenced morphological traits to different extents. It was found that the hormones should be applied in accordance with the trait. For example, when a high rooting percentage is required, it is better to apply 1000 mL NAA. When a high number of roots are intended, 1000 mL NAA and 200 mL SA should be applied, and when we want to grow long roots, 1000 mL NAA or 100 µM ABA should be applied.

IBA has usually a commercial use for rooting. Other commercially used auxins are IAA and NAA. Many chemical analogs have been synthesized and their activity as auxin has been investigated (Jönsson, 1961), but none of them has ever employed for rooting at large scale. The differences observed in the effect of different auxins may be related to the nature of the compound, the tendency to receive auxin in rooting, and/or the concentration of free auxin reaching target cells. Also, it depends on several factors including the uptake, mobilization, and conversion of the applied compound as well as the quantity of auxin synthesized by the plant itself (Hartmann et al., 1997).

Conclusion

We found that hormone application had a positive effect on rooting percentage. These results may be helpful for commercial production of *S. arboricola*. Leaf-containing 10 cm cuttings were used for this study and obtained a very high rooting percentage. It was revealed that the application of different hormones influenced morphological traits to varying degrees. According to the results, certain hormones should be applied for each trait. For example, when a high rooting percentage is required, it is better to apply 1000 mL NAA. When a high number of roots are intended, 1000 mg L^{-1} NAA and 200 mL SA should be applied, and when we want to grow long roots, 1000 mg L^{-}1 NAA or 100 µM ABA should be applied. The highest catalase enzyme activity was observed in cuttings treated with 1000 mg L^{-1} IBA + 50 µM ABA + 200 mL L^{-1} SA (T_{11}) followed by those treated with 200 mL L^{-1} SA (T_8), 300 mL L^{-1} SA (T_9), and 1000 mg L^{-1} NAA (T_4).

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