ZIBELINE INTERNATIONAL

ISSN: 2521-2931 (Print)
ISSN: 2521-294X (Online)
CODEN : MJSAEJ

Malaysian Journal of Sustainable Agriculture (MJSA)

DOI : http://doi.org/10.26480/mjsa.02.2019.01.04

CrossMark

RESEARCH ARTICLE

STUDY ON THE ROOTING OF PICEA ABIES CUTTINGS UNDER AUXINS, SUBSTRATES AND BOTTOM HEAT

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ARTICLE DETAILS

Article History:

Received 15 November 2018
Accepted 17 December 2018
Available online 15 January 2019

ABSTRACT

The effect of rooting hormones, substrate and bottom heat was studied on the rooting of the cuttings of *Picea abies*. The treatments included bottom heat at two levels, hormones at six levels and rooting substrate at three levels. The recorded traits included rooting percentage, the number of roots; root length and root dry weight. It was found that the applied hormones had no considerable effect on rooting and the recorded traits, so that the application of 2000 and 4000 mg/l IBA had no significant difference with no-hormone application on all three substrates with or without the use of bottom heat. Cuttings treated with NAA produced no roots in any of the studied three substrates. The highest number of roots was produced under the treatment of sand + perlite × 4000 mg/l IBA × no-bottom heat. The treatment of no-bottom heat × no-hormone × perlite produced the longest root. The highest root dry weight was devoted the treatments of no-bottom heat × no-hormone × sand and no-bottom heat × 2000 mg/l IBA× sand.

KEYWORDS

auxin, bottom heat, *Picea abies* (L.), rooting

1. INTRODUCTION

Norway spruce (*Picea abies* (L.) Karst 'Nidiformis') is a short conifer with needle-like leaves similar to cushion from the family of Pinaceae which is usually grown as an ornamental plant in gardens and parks [1]. It is propagated by seeds and cuttings. But since propagation by cutting is the easiest, cheapest and best method of producing true-to-type plants, it is recommended to propagate by cutting. It is known that semi-hardwood cuttings of spruce hardly start rooting and so, it is very important to understand the most appropriate conditions for the rooting of this species to maximize their rooting [2]. The rooting of cuttings, especially hardly rooting cuttings, is influenced by various environmental and internal factors including the preparation of the cutting at the right time during plant growth cycle, the use of proper rooting substrate, the optimum temperature and moisture in the rooting medium, and the application of growth regulators with correct dose; so, these factors must be taken care of to obtain acceptable rooting rate [3,4]. One of the environmental factors that affect the rooting of the cuttings, especially hard-to-root cuttings, is to use bottom heat. It is reported that bottom heat at rooting substrate can activate cofactors and initiate rooting [5,6].

Rooting substrate is an important factor in the rooting of hard-to- root cuttings; the selection of appropriate substrate increases the efficiency of rooting induction. The lack of appropriate planting substrate can decrease water uptake efficiency, cause plant wilting and reduce cell size. It seems that the use of appropriate planting substrate is an important step in the propagation of horticultural crops which can improve rooting rate and the number of rooted cuttings per unit area. The simplest, widely used rooting medium for cuttings is sand. Sand is the heaviest and most common rooting substrate for cuttings. It lacks nutrients and has a high rate of aeration. The rooting of leafy cuttings is reported to be increased in sandy substrate [7]. A researcher reported sandy substrate as the most appropriate substrate for the rooting of semi-hardwood cutting of *Callistemon viminalis* [8]. Perlite is a rooting substrate that is composed of gray-while silicon with mineral and volcanic origins. Perlite can absorb and hold water 3-4 times as much as its weight. It has a great drainage, but it lacks nutrients and cation exchange property [9]. A previous researcher reported that the application of perlite mixed with other substrates like peat can improve the growth and development of the roots [10].

Another factor that can improve the rooting of the cuttings is the rate of hormones and growth regulators. Among these factors, the highest effect is observed to be of auxins [11]. IBA and NAA are the most common auxins used for the rooting of cuttings. The application of auxins for most tree species increases the rooting and improves root quality. Auxins stimulate the formation of roots on cuttings by stimulating cellular division and activating rhizokalin [12]. Today, it is well accepted that the adventitious roots on stems are initiated by natural or application of synthetic auxins and in fact, the rate of rooting on stems is proportional to the rate of auxin application. Based on a study, the number of roots was significantly influenced by the interaction between cutting and growth regulators [13]. The rooting of the cuttings of Jojoba and Guava by the application of IBA is reported. It is argued that auxin improves cellular division, which in turn, results in a higher number of roots [14,15].

Spruce is hard-to-root. Consequently, the objective of the present study was to examine the effect of synthetic auxins and different rooting substrates as well as bottom heat on the rooting of the cuttings of Norway spruce (*Picea abies* (L.) Karst 'Nidiformi').

2. MATERIALS AND METHODS

2.1 Trial design

The study carried out as a factorial experiment based on a Randomized Complete Block Design with three factors. The first factor was bottom heat [including the lack of bottom heat (a_1) and the application of bottom heat (a_2)], the second factor was the application of rooting hormones [no hormone (b_1), 2000 mg/l IBA (b_2), 4000 mg/l IBA (b_3), 2000 mg/l NAA (b_4), 4000 mg/l NAA (b_5), 2000 mg/l IBA + 2000 mg/l NAA (b_6)] and third factor was rooting substrate [sand (c_1), perlite (c_2), sand + perlite (v/v) (c_3)] at three replications in a tunnel plastic greenhouse at 18-20°C equipped with bottom heat and mist systems.

2.2 Plant materials

The leafy cuttings of Norway spruce with the length of 8-12 cm and a diameter of 1.5-2.5 mm were obtained from an ornamental propagator company in Ramsar, Iran in February 2014. Then, the final 2 cm of the cuttings was soaked in the hormone solution for 10 seconds. After that, they were weathered for about 10 seconds for the alcohol of the solution to vaporize. Afterwards, they were planted in rooting substrates. Before planting, all substrates were disinfected with copper oxychloride. The cuttings were daily irrigated with mist system. The bottom heat was adjusted at 22-24°C during winter and it was stopped in early spring.

2.3 Experimental traits

The cuttings were taken from the substrate 140 days after planting date to record the studied traits. The recorded traits included rooting percentage, the number of roots per cutting and the length of the longest root. To measure the dry weight of the roots, they were parted from the cuttings and weighed. Then, they were oven-dried at 105°C for 24 hours to estimate their dry weight. The data were analyzed by the MSTATC statistical software package and the means were compared with LSD test.

3. RESULTS

3.1 Rooting percentage

Means comparison of the data of rooting percentage revealed that the highest rooting was related to the treatments of no hormone and different rates of IBA at three substrates and in both with and without bottom heat. The highest rooting percentage (99%) was observed in the treatments of no bottom heat × no hormone × sand, no bottom heat × 2000 mg/l IBA × sand, no bottom heat × 4000 mg/l IBA × perlite, no bottom heat × 4000 mg/l IBA × sand, bottom heat × no hormone × perlite, and bottom heat × no hormone × perlite. Different NAA concentrations had no effect on the rooting of the cuttings and in total, NAA had no specific effect on the rooting of Norway spruce (Table 1).

Table 1: Means comparison of the effect of different treatments on the measured traits

	Rooting percentage	Number of roots	Root length (cm)	Root dry weight (g)
No bottom heat + no hormone + sand	99 a	4.00 bcd	4.25 cde	0.036 d-h
No bottom heat + no hormone + perlite	88 a	2.66 def	6.19 ab	0.04 d-h
No bottom heat + no hormone + sand + perlite	88 a	4.66 a-d	6.14 ab	0.096 b
No bottom heat + 2000 mg/l IBA + sand	99 a	5.66 abc	2.04 ghi	0.023 f-i
No bottom heat + 2000 mg/l IBA + perlite	88 a	4.33 bcd	6.22 ab	0.063 b-e
No bottom heat + 2000 mg/l IBA + sand + perlite	66 a	5.33 abc	3.37 c-g	0.046 d-g
No bottom heat + 4000 mg/l IBA + sand	99 a	4.33 bcd	2.94 e-h	0.04 d-h
No bottom heat + 4000 mg/l IBA + perlite	99 a	4.3 bcd	4.70 b-e	0.043 d-h
No bottom heat + 4000 mg/l IBA + sand + perlite	88 a	7.00 a	4.90 bc	0.07 bcd
No bottom heat + 2000 mg/l NAA + sand	0 a	0.00 g	0.00 j	0.00 i
No bottom heat + 2000 mg/l NAA + perlite	0 a	0.00 g	0.00 j	0.00 i
No bottom heat + 2000 mg/l NAA + sand + perlite	0 a	0.00 g	0.00 j	0.00 i
No bottom heat + 4000 mg/l NAA + sand	0 a	0.00 g	0.00 j	0.00 i
No bottom heat + 4000 mg/l NAA + perlite	0 a	0.00 g	0.00 j	0.00 i
No bottom heat + 4000 mg/l NAA + sand + perlite	0 a	0.00 g	0.00 j	0.00 i
No bottom heat + 2000 mg/l IBA + 2000 mg/l NAA + sand	33 a	1.33 fg	1.45 hij	0.016 ghi
No bottom heat + 2000 mg/l IBA + 2000 mg/l NAA + perlite	0 a	0.00 g	0.00 j	0.00 i
No bottom heat + 2000 mg/l IBA + 2000 mg/l NAA + sand + perlite	0 a	0.00 g	0.00 j	0.00 i
Bottom heat + no hormone + sand	88 a	4.33 cde	4.32 cde	0.136 a
Bottom heat + no hormone + perlite	99 a	5.66 abc	7.88 a	0.06 cde
Bottom heat + no hormone + sand + perlite	88 a	4.33 bcd	4.91 bc	0.093 bc
Bottom heat + 2000 mg/l IBA + sand	99 a	5.33 abc	4.11 cde	0.136 a
Bottom heat + 2000 mg/l IBA + perlite	77 a	5.66 abc	4.85 bcd	0.053 def
Bottom heat + 2000 mg/l IBA + sand + perlite	44 a	4.66 a-d	3.31 c-g	0.066 b-e
Bottom heat + 4000 mg/l IBA + sand	66 a	6.33 ab	3.95 c-f	0.063 b-e
Bottom heat + 4000 mg/l IBA + perlite	44 a	3.66 cde	2.30 f-i	0.043 b-h
Bottom heat + 4000 mg/l IBA + sand + perlite	44 a	2.66 def	3.05 d-h	0.033 e-i
Bottom heat + 2000 mg/l NAA + sand	0 a	0.00 g	0.00 j	0.00 i
Bottom heat + 2000 mg/l NAA + perlite	22 a	0.66 fg	0.00 j	0.01 hi
Bottom heat + 2000 mg/l NAA + sand + perlite	0 a	0.00 g	0.00 j	0.00 i
Bottom heat + 4000 mg/l NAA + sand	0 a	0.00 g	0.00 j	0.00 i
Bottom heat + 4000 mg/l NAA + perlite	0 a	0.00 g	0.00 j	0.00 i
Bottom heat + 4000 mg/l NAA + sand + perlite	0 a	0.00 g	0.00 j	0.00 i
Bottom heat + 2000 mg/l IBA + 2000 mg/l NAA + sand	0 a	0.00 g	0.00 j	0.00 i
Bottom heat + 2000 mg/l IBA + 2000 mg/l NAA + perlite	0 a	0.00 g	0.00 j	0.00 i
Bottom heat + 2000 mg/l IBA + 2000 mg/l NAA + sand + perlite	33 a	2.66 def	1.03 ij	0.036 d-h

Means followed by the same letters in each column are not significantly different in LSD test.

3.2 The number of roots

According to means comparison of root number data, the treatment of no bottom heat + 4000 mg/l IBA × sand × perlite produced the highest number of roots (7 roots) followed by the treatment of bottom heat × 4000 mg/l IBA × sand with 6.33 roots. No roots were produced under the application of NAA at all three substrates with and without bottom heat as well as the treatments of no bottom heat × 2000 mg/l IBA + 2000 mg/l NAA × perlite, no bottom heat × 2000 mg/l IBA + 2000 mg/l NAA × sand + perlite, bottom heat × 2000 mg/l IBA + 2000 mg/l NAA × sand and bottom heat × 2000 mg/l IBA + 2000 mg/l NAA × perlite. Among the treatments which resulted in the growth of roots, the lowest number of roots was observed in the treatment of bottom heat × 2000 mg/l NAA × perlite (Table 1).

3.3 Root length

Means comparison of root length indicated that the longest roots (7.88 cm) were produced under the treatment of bottom heat × no hormone × perlite followed by the treatment of no bottom heat × 2000 mg/l IBA × perlite with the root length of 6.22 cm. After the treatments in which no roots were produced and so, the root length was zero (Table 1), the shortest roots were related to bottom heat × 2000 mg/l IBA + 2000 mg/l NAA × sand + perlite with the length of 1.03 cm followed by the treatment of no bottom heat × 2000 mg/l IBA + 2000 mg/l NAA × sand with the root length of 1.45 cm (Table 1).

3.4 Root dry weight

Means comparison showed that the treatments of bottom heat × no hormone × sand and bottom heat × 2000 mg/l IBA × sand resulted in the highest root dry weight (0.136 g). The lowest root dry weight was related to the treatment of NAA at three substrates with and without bottom heat. Also, the root dry weight was zero in the treatments of bottom heat × 2000 mg/l IBA + 2000 mg/l + perlite, bottom heat × 2000 mg/l IBA + 2000 mg/l NAA × sand, no bottom heat + 2000 mg/l IBA + 2000 mg/l NAA + sand + perlite, and no bottom heat + 2000 mg/l IBA + 2000 mg/l NAA × perlite. Besides, the treatments of bottom heat × 2000 mg/l NAA × perlite and no bottom heat + 2000 mg/l IBA + 2000 mg/l NAA × sand produced the lowest root dry weights, respectively (Table 1).

4. DISCUSSION

As mentioned in the Results section, cuttings which were not treated with hormones had better rooting than those treated with hormones in all three substrates with or without bottom heat. It implies that the cuttings of Norway spruce prepared in February did not need hormone treatments for rooting and can readily initiate rooting in sand or perlite under mist system. A researcher argued that semi-hardwood cuttings have an optimum auxin level and that the increase in exogenous hormone concentration disrupts the plant hormone balance, resulting in the loss of rooting which is consistent with our results.

Among IBA and NAA treatments, the treatment of 2000 mg/l IBA had the second highest rooting after control. The treatments of NAA and IBA + NAA exhibited the lowest rooting. These results are in agreement with a researcher who found that IBA was more effective than NAA in rooting and another researcher who found that IBA was superior over other auxins in successful rooting of different types of cuttings [16,17].

The failure of hormones in inducing roots of Norway spruce cuttings as compared to control can be related to some factors, for example inappropriate concentrations of the hormones which can be confirmed only by an experiment on a much wider range of hormone concentrations. However, it should also be noted that the treatment of auxin is not effective on the rooting of all species. The application of NAA alone or combined with rooting substrate and bottom heat system had a negative effect on the rooting of Norway spruce so that the increase in its concentration not only did not increase rooting but also decreased. This result is consistent with a researcher who believe that high auxin concentrations can be harmful to the tissue of the bottom of the cuttings and Arteca (1997) who argued that NAA is stronger and more stable than natural auxins and so, it is better to be used in lower concentrations [18,19].

It is known that rooting percentage and the quality of cuttings directly depend on planting substrate in most cases [20]. Many researchers suggest that the use of substrate with neutral pH like sand and perlite improves rooting of the cuttings. In the present study too, sand and perlite increased the rooting. A researcher reported that IBA and sand + perlite substrates were effective on the rooting of the cuttings of *Thuja occidentalis* [21]. Qasemi *et al.,* (2013) also, suggested sand as the best substrate for the rooting of semi-hardwood cuttings of guava and stated that a substrate with neutral pH and improved drainage increased the rooting which is in agreement with our results [22].

One advantage of auxin is reportedly the increase in the number of roots per cutting [23]. It is believed that auxin increases the number of roots through stimulating the growth of adventitious roots and the development of latent and preformed root initiators [24]. However, it should be noted that the treatment of NAA adversely impacted the number of roots in the present study. The normal number of roots in control implies that the control cuttings had indigenous auxin required for optimum production of roots. A scholar believes that 1-2% IBA increased the number of roots in magnolias cuttings and very high concentrations of NAA decreased it, which is consistent with our results [25].

In a study on rooting of guava stem cuttings, a researcher showed that the longest roots and the highest number of roots were observed in the treatment of 1000 mg/l IBA and NAA [26]. The highest rooting of jojoba and guava cuttings was obtained under IBA treatments. They believed that auxin improved cellular division resulting in a higher number of roots.

Finally, it can be concluded that although hormone treatments had no significant impact on the rooting percentage of Norway spruce, they improved the production of roots alone or in combination with other treatments like rooting substrate.

The highest root length was obtained under the treatment of no hormone on perlite with bottom heat. The treatments of IBA were more appropriate for the increase in root length than NAA. The increase in the root length under the application of growth regulators can be caused by higher hydrolysis of carbohydrates. By increasing metabolism in their application spot and synthesis of new proteins, auxins contribute to the development and division of cells, which in turn, results in higher numbers and length of roots [27,28]. The increase in root length under the application of 1000 mg/l IBA is reported for *Poinsettia pulcherrima*, too [29]. A researcher reported that the best treatment for rooting and root length was 2000 mg/l IBA + sand [30]. The highest root length of *Thuja occidentalis* was found under perlite and 2000 mg/l IBA.

It was found that root dry weight was the highest under the application of bottom heat on sand substrate without the application of hormone and with the application of 2000 mg/l IBA. According to a research, the application of high concentrations of IBA positively influenced the quality and dry weight of roots in carnation and chyrsanthemum, which is inconsistent with our results. However, he reported that different concentrations of NAA had no significant effect on this process which is in agreement with our results.

A researcher obtained the highest root weight (1.12 g) from the treatment of NAA. In a study on the effect of auxin and different substrates on rooting of *Thuja occidentalis*, another researcher stated that the highest root dry weight was produced under the treatment of perlite × 4000 mg/l IBA. Based on a research, different substrates and hormone concentrations had a positive effect on the dry weight of *Dodoneae viscosa* L. roots [31-34].

5. CONCLUSION

In the present study, it was revealed that the highest rooting of of *P. abies* (L.) Karst ‘Nidiformis’ cuttings was obtained under the treatment of no hormones and sand or perlite substrates. Therefore, it can be concluded that the appropriate substrate and environmental factors are more important for the rooting of cuttings of Norway spruce than internal factors. In fact, the hard- to- root feature of this plant cuttings is not correlated to the deficiency of auxin in the stem tissues; although further studies are required to confirm this hypothesis.

ACKNOWLEDGEMENTS
Financial support by Rasht Branch, Islamic Azad University Grant No. 4.5830 is gratefully acknowledged.

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Cite The Article: Shahram Sedaghathoor, Somayeh Abdizadeh Sarem (2019). Study On The Rooting Of Picea Abies Cuttings Under Auxins, Substrates And Bottom Heat. *Malaysian Journal of Sustainable Agriculture*, 3(2): 01-04.