INFLUENCE OF NAPHTHALENE ACETIC ACID (NAA) ON SPROUTING AND ROOTING POTENTIAL OF STEM CUTTINGS OF BOUGAINVILLEA

Noor-un-nisa Memon[1*], Noman Ali[1], Muhammad Ayub Baloch[1], Qammaruddin Chachar[2]
Department of Horticulture, Sindh Agriculture University, Tandojam, Sindh, Pakistan[1]
Department of Crop Physiology, Sindh Agriculture University, Tandojam, Sindh, Pakistan[2]
Corresponding e-mail: **norimemon@gmail.com**

ABSTRACT: *An experiment was conducted to assess the effect of various concentrations of Naphthalene acetic acid (NAA) on sprouting and rooting potential of Bougainvillea stem cuttings. The stem cuttings of two different variegated varieties viz. white and red about 6 inches in length were obtained from three year old plants. The basal ends of the cuttings were dipped in NAA solutions viz. 2000, 4000 and 6000 mg L^{-1} plus control treatment by quick dip method for 10 seconds before planting them in growing medium. The results revealed that all the traits related with sprouting and rooting potential of stem cuttings of Bougainvillea were significantly influenced by various NAA concentrations, while varieties did not influence these characters significantly with the exception of number of sprouts per cutting, sprouting and rooting percentage. The best results obtained from the treatment where NAA was applied at the concentration of 6000 mg L^{-1} with 3.133 sprouts per cutting, 78.33 sprouting percentage, 0.680 cm length of sprouts, 75.00 rooting percentage, 13.862 cm length of roots, 27.833 roots per cutting, 28.20 leaves per cutting and 6.007 cm length of the leaves. Correlation matrix exhibited positive association between sprouting and rooting related parameters of Bougainvillea stem cuttings except length of the sprouts.*

INTRODUCTION

Bougainvillea is a lush evergreen subtropical vine. It has a spreading, round plant habit with a height and spread of up to 20 feet [1]. It is used in mass plantings, as shrubs or bushes, and as ground cover [2]. The plant is also used as hedges, barriers, and slope coverings. For large, difficult-to-maintain areas, Bougainvillea is an excellent ground cover. It can cover a whole hillside and will choke out weed growth. *Bougainvillea*, belongs to the family Nyctaginaceae, with five species include *Bougainvillea glabra***,** *Bougainvillea buttiana***,** *Bougainvillea peruviana***,** *Bougainvillea spectabilis* and *Bougainvillea spinosa***.** The flower of the plant is small and generally white, but each cluster of three flowers is surrounded by three or six bracts with the bright colours associated with the plant, including pink, magenta, purple, red, orange, white, or yellow. *Bougainvillea glabra* is sometimes referred to as "paper flower" because the bracts are thin and papery. The fruit is a narrow five-lobed achene [1].

The general practice of multiplication for most of the perennial ornamental plants is by the use of vegetative plant parts including stem, leaves, terminal buds and roots due to its simplicity and practicability in developing countries [3]. Vegetative propagation of ornamental plants through stem cutting is one of the cheapest and sometimes, the only method available for multiplication. However, under normal conditions, wide variability is noticed in different cultivars of the same species; while some cultivars root easily, others are either difficult or fail to root. In Bouganvillea the success of propagation by stem cutting is very limited. Under normal conditions mostly growers observed poor rooting percentage. However the rooting responses vary from variety to variety. For example the varieties like Thimma and Dr. Rao which are difficult to root and therefore need proper handling and careful manipulation of rooting conditions.

It is well known that the success of rooting of the woody stem cuttings, in the majority of ornamental plants and fruit trees depends mainly on the physiological stage of the mother plant [4], the time of planting of the cutting [5,6] and the type of growth regulators used [7].

Treatment of cuttings for ease in rooting is an old horticultural practice. In the early historic days, people used to treat cuttings with manganese, boron and phosphorus but with the discovery of auxins in 1934 for their simulative effects on root initiation was a major milestone in the history of plant propagation. Various auxins such as Indole Acetic Acid (IAA), Indole Butyric Acid (IBA), Naphthalene Acetic Acid (NAA) and 2,4-Dichloropheoxy Acetic Acid (2,4-D) have been reported to promote rooting in cuttings of the most of the plant species. Each auxin's concentration varies from plant to plant and type of the cuttings used. IBA or NAA or combination of both is mostly recommended for rooting of cuttings. These are available in liquid, talc, tablet, and gel formulations. Liquid formulations are generally sold as solvent based concentrates that may be diluted to the desired concentration for treating cuttings of specific crops [8]. However 2,4-D is a potent weed killer and it may inhibit shoot development in certain species. These compounds are insoluble in water and can be dissolved in a few drops of alcohol or ammonium hydroxide, before adding water. Cuttings of some species root readily without an auxin treatment, while cuttings of other species benefit from auxin treatment through enhanced promotion of rooting; benefits may be dependent upon the species and cultivar, condition of the cutting wood, time of year, and other factors [9].

Commercial root-promoting chemicals are normally applied to the basal portion of cuttings using a liquid or talc formulation of auxin. The quick-dip method is often preferred by commercial propagators for application of liquid auxin formulations for reasons of economy, speed, ease, and uniformity of application and results. An extended basal soak may be utilized for some difficult-to-root species [10].

Realizing the role of auxins in rooting initiation and development, the present study was therefore designed to investigate the effect of NAA on sprouting and rooting responses using stem cuttings of *Bouganvillea*.

MATERIAL AND METHODS

In order to investigate the influence of NAA on sprouting and rooting potential of *Bougainvillea* stem cuttings, the experiment was conducted during the year 2012. The stem cuttings of two different colour flower varieties (white and red) about 6 inches in length were obtained from three year old plants during March, 2012. The stem cuttings of two different variegated varieties about 6 inches in length were obtained from three year old plants of Bougainvillea. While preparing the cuttings, a smooth cut in each cutting was given on distal end and slanting cut was given at lower end just below the node. The basal ends of the cuttings were dipped in NAA solutions viz. 2000, 4000 and 6000 mg L^{-1} plus control treatment with no NAA by quick dip method for 10 seconds before planting them in the rooting medium. The solutions were prepared by dissolving the NAA compound in 95% ethanol and adding distilled water. The growing medium was prepared by mixing equal ratio of sand, farm yard manure and garden soil in the ratio of 1:1:1 (v/v). This growing medium was analyzed for soil texture by Bouyoucos Hydrometer method, pH of 1:5 soil-water extract by pH meter, EC of 1:5 soil-water extract by EC meter and organic matter by Walkley-Black method. The growing medium of the cuttings was sandy loam in texture with 75% sand, 16% clay and 7.5% silt content. The soils were non-saline with EC 1.98 dS/m and alkaline in nature with pH 7.8. The organic matter content of the growing medium was 0.75%. The mixture of growing medium was filled in perforated white polythene bags of half kg, tightly leaving one inch space at the top. One cutting was planted in each polythene bag. The experiment was conducted in a three replicated Completely Randomized Design (CRD) each treatment was comprised of 10 cuttings and a total of 120 cuttings of each variety were managed. The data on number of sprouts per cutting, sprouting percentage per treatment, length of sprouts per cutting, rooting percentage of treatments, length of roots per cutting, number of roots per cutting, number of leaves per cutting and the length of the leaves were recorded after two months of plantation. The data thus obtained were statistically analyzed using Analysis of variance (ANOVA) and means were compared according to Duncan's Multiple Range Test [11]. The correlation studies between sprouting and rooting related parameters were also evaluated.

RESULTS

The results revealed that all the traits related with sprouting and rooting potential of stem cuttings of *Bougainvillea* were significantly influenced by various NAA concentrations, while varieties did not influence these characters significantly with the exception of number of sprouts per cutting, sprouting and rooting percentage. However, the interaction between varieties and NAA concentrations was also observed at par for all these parameters.

The data in Table 1 depicts that sprouting percentage (78.33%) of stem cuttings was observed higher in cuttings which are treated with the highest NAA concentration of 6000 mg L^{-1}. These results are statistically similar with the results of 68.33 for sprouting percentage obtained from the treatment where NAA was applied at the rate of 4000 mg L^{-1}. However, the sprouting percentage was lowest (26.67%) in control, where cuttings were not treated with NAA. Varieties had no significant differences for sprouting percentage. Mean number of sprouts per cutting ranged from 1.733 to 3.133. All NAA concentrations exhibited statistically similar number of sprouts per cutting with the maximum number of sprouts of 3.133 per cutting from the treatment where NAA was applied @ 6000 mg L^{-1}. There was a simultaneous improvement in the number of sprouts per cutting with increasing NAA concentrations and showing the significance over the control. The lowest number of sprouts per cutting was recorded from the control where no NAA was applied. On the basis of varietal comparison, white colored variety significantly had more number of sprouts of 2.95 as compared to red colored variety (2.283). Regarding length of the sprouts, maximum mean length of sprouts per cutting (0.680 cm) was recorded from the cuttings where treated with the highest NAA concentration of 6000 mg L^{-1}. However these results are significantly reduced from 0.680 to 0.462 with the decreasing levels of NAA i.e from 6000 to 2000 mg L^{-1}. The lowest length of sprouts per cutting of 0.383 cm was noted in control. White variety exhibited greater length of sprouts per cutting (0.528 cm) as compared to red coloured variety (0.505 cm) but statistically there had no varietal differences.

Table 2 shows that the trend of rooting percentage was observed increased with the increasing concentrations of NAA i.e 2000 to 6000 mg L^{-1}. Maximum rooting percentage of 75% was observed from the NAA treatment where higher concentration of NAA (6000 mg L^{-1}) was applied. However these results were at par with the results of 68.33% obtained from the treatment where NAA was applied at the rate of 4000 mg L^{-1}. Control treatment had statistically significant differences with NAA treatments and obtained minimum rooting percentage of 20%. On the basis of varietal comparison, white colored variety had significantly maximum rooting percentage of 60 as compared to red colored variety (48.33). Maximum number of roots (27.833) was observed from the treatment where NAA concentration was applied at the rate of 6000 mg L^{-1} followed by the results of 15.333 in response to the treatment where NAA was applied at the rate of 4000 mg L^{-1}. Control had minimum number of roots (4.867) per cutting followed by 11.300 in response to the treatment where NAA was applied at the rate of 2000 mg L^{-1}. Comparatively greater number of roots per cutting (15.450) was observed in White variety as compared to the red variety (14.317). This indicates that the impact of increasing NAA concentrations increased the number of roots per cutting remarkably. Maximum length of the roots (13.862 cm) was also observed from the treatment where higher concentration of NAA i.e. 6000 mg L^{-1} was applied. However rest of two NAA concentrations i.e. 2000 and 4000 mg L^{-1} exhibited statistically similar results of 9.278 and 9.695 cm respectively. These results are also at par with the results obtained from control treatment (8.802 cm).

Table 3 depicts that the maximum number of leaves (28.2) per cutting was observed from the treatment where NAA was applied at the concentration of 6000 mg L^{-1}. These results are significantly different from the results of 20.267

Table 1. **Effect of various concentrations of Naphthalene acetic acid (NAA) on sprouting potential of stem cuttings of Bougainvillea.**

NAA (mg L^{-1})	Sprouting (%) per NAA concentration			No. sprouts/cutting			Length of the sprouts/cutting		
	White	Red	Mean	White	Red	Mean	White	Red	Mean
Control (No NAA)	33.33	20.00	**26.67 C**	2.067	1.400	**1.733 B**	0.360	0.407	**0.383 D**
2000	60.000	50.00	**55.00 B**	3.067	2.467	**2.767 A**	0.483	0.440	**0.462 C**
4000	76.67	60.00	**68.33 AB**	3.000	2.667	**2.833 A**	0.583	0.500	**0.542 B**
6000	83.33	73.33	**78.33 A**	3.667	2.600	**3.133 A**	0.687	0.673	**0.680 A**
Mean	**63.33 A**	**50.83 B**		**2.950 A**	**2.283 B**		**0.528**	**0.505**	

Table 2. **Effect of various concentrations of Naphthalene acetic acid (NAA) on rooting potential of stem cuttings of Bougainvillea.**

NAA (mg L^{-1})	Rooting (%) per NAA concentration			No. roots/cutting			Length of the roots/cutting		
	White	Red	Mean	White	Red	Mean	White	Red	Mean
Control (No NAA)	30.00	10.00	**20.00 C**	5.933	3.800	**4.867 D**	9.570	8.033	**8.802 B**
2000	56.67	50.00	**53.33 B**	10.067	12.533	**11.300 C**	9.910	8.647	**9.278 B**
4000	76.67	60.00	**68.33 A**	15.867	15.200	**15.533 B**	9.323	10.067	**9.695 B**
6000	76.67	73.33	**75.00 A**	29.33	25.733	**27.833 A**	13.273	14.450	**13.862 A**
Mean	**60.00 A**	**48.33 B**		**15.450**	**14.317**		**10.519**	**10.299**	

Table 3. **Effect of various concentrations of Naphthalene acetic acid (NAA) on number of leaves and length of the leaves sprouted from stem cuttings of Bougainvillea.**

NAA (mg L^{-1})	No. of leaves per cutting			Length of the leaves/cutting		
	White	Red	Mean	White	Red	Mean
Control (No NAA)	11.667	12.167	**11.917 C**	3.65	2.347	**2.998 B**
2000	19.733	19.000	**19.367 B**	3.03	2.628	**2.829 B**
4000	20.667	19.867	**20.267 B**	5.157	5.406	**5.282 A**
6000	31.667	24.733	**28.200 A**	5.132	6.881	**6.007 A**
Mean	**20.933**	**18.942**		**4.242**	**4.316**	

Table 4. Correlation matrix between sprouting and rooting parameters.

SPROUTING/ ROOTING PARAMETERS	Sprouts/ cutting	Length of sprouts	Length of roots	Number of roots	Number of Leaves	Length of leaves
Sprouts/cutting	-					
Length of sprouts	-0.207^{NS}	-				
Length of roots	0.451^{*}	-0.045^{NS}	-			
Numberof roots	0.655^{**}	-0.149^{NS}	0.720^{**}	-		
Number of Leaves	0.687^{**}	-0.069^{NS}	0.681^{**}	0.872^{**}	-	
Length of leaves	0.264^{NS}	-0.168^{NS}	0.536^{**}	0.643^{**}	0.417^{*}	-

and 19.367 observed from the treatments where NAA was applied at the concentration of 4000 and 2000 mg L^{-1} respectively. However minimum number of leaves of 11.917 was obtained from the control treatment followed by 19.367 from the treatment where NAA was applied at the rate of 2000 mg L^{-1}. Better length of the leaves of 6.007 cm was observed from the treatment where highest concentration i.e 6000 mg L^{-1} was applied. These results were at par with the results of 5.282 cm obtained from the reduced concentration of NAA (4000 mg L^{-1}). The lowest concentration of NAA (2000 mg L^{-1}) and control exhibited statistically similar results of 2.829 and 2.998 respectively.

CORRELATION STUDIES

Correlation matrix in Table 9 exhibited that number of sprouts was positively associated with length of the roots, number of roots, number of leaves and length of the leaves with "r" value of 0.451, 0.655 and 0.687 respectively but negatively associated with length of the leaves (r = 0.264) and length of the sprouts (r = -0.207). However length of the sprouts was also negatively correlated with all other parameter related with roots and leaves including length of the roots, number of roots, number of leaves and length of the leaves.

Length of the roots was positively correlated with number of roots (r=0.720), number of leaves (r=0.681) and length of the leaves (r=0.536). However number of leaves (r=0.872) and length of the leaves (r=0.643) had also positive association with number of roots and with each other (r=0.417).

DISCUSSION

Rooting and sprouting potential of stem cuttings is one of the critical steps in plant propagation of woody plants. It varies from specie to specie including so many other factors. However propagation through seeds cause genetic variability and makes the crops prone to different diseases. Vegetative propagation has an advantage in developing true to type, disease free varieties of economically and commercially important plants [12]. Vegetative propagation of Bougainvillea by stem cuttings has been found to be very effective because of its simplicity and practicability in our developing countries. However the rooting rate of success is very low and varies from specie to specie. Due to limited rates of success in sprouting and rooting, many researchers tried various auxins for initiation of rooting in cuttings of various horticultural crops as reported by [13,14].

The present study was undertaken to investigate the effect of auxin i.e. Naphthalene acetic acid (NAA) on sprouting and rooting potential of stem cuttings of *Bougainvillea* of two different varieties. The results obtained on various parameters viz., number of sprouts per cutting, sprouting percentage per treatment, length of sprouts per cutting (cm), rooting percentage per treatment, length of roots per cutting (cm), number of roots per cutting, number of leaves per cutting and length of leaves per cutting (cm) are under discussion in this chapter.

In present study auxin (NAA) treated cuttings had more potential for number of sprouts per cutting as compared to control treatment. These results are in accordance with the results of [2]. They reported more number of sprouts from November planted stem cuttings of Bougainvillea variety Torch glory treated with auxin (IBA) at the concentration of 3000 mg L^{-1}. However they also observed the lowest number of sprouts with the increasing levels of IBA upto 4500 mg L^{-1} in the same Bougainvillea variety Torch glory. Awad *et al*. [15] reported higher percentage of bud sprouts from the Bougainvillea stem cuttings of 12 mm thickness treated with auxin at the concentration of 6000 ppm of Indole butyric acid (IBA). Kochhar *et al*. [12] also observed maximum number of sprouts from stem cuttings of *Jatropha curcas* when treated with auxin i.e NAA @ 100 ppm as compared to 10 ppm. This concentration of NAA is much lower than the concentration we applied in our study. This may be the cause due to different type of plant, season variability, and thickness of the stem cutting. Wahab *et al*. [16] reported better sprouting from the semi hard wood cuttings of guava when treated with NAA @ 2000 ppm. They also reported that sprouting is mainly attributed to the stored carbohydrates in the cuttings used for sprouting. However with auxin application to the cutting and subsequent increase in the rooting may result in the increase of sprouting, this indirect effect of auxin on sprouting highlights the role of certain materials produced in the roots, responsible for sprouting. Jadhav [17] reported maximum number of sprouts and length of sprout from stem cuttings of phalsa treated with NAA @ 200 ppm.

In present study, maximum percentage of sprouting was also observed from the treatments where NAA was applied at higher concentration (6000 mg L^{-1}). Singh *et al*. [2] obtained 100% sprouting from January planted stem cuttings of Bougainvillea treated with various concentrations of IBA including 2000, 2500 and 3000 mg L^{-1}. However, Tiwari and Das [18] observed better response for sprouting on much reduced levels of NAA (1000 ppm). This may be the cause due to different type of plants and may be due to diameter of the stem cutting. They propagated two shrub medicinal species viz *Caesalpinia bonduc* and *Embelia tsjeriam* through stem cuttings by applying exogenous plants growth regulators including Indole Acetic Acid (IAA), α- Naphthalene Acetic Acid (NAA) and Indole 3 Butyric Acid (IBA). Majeed and Mughal [19] reported 75% sprouting from the stem cuttings of *Aesculus indica* treated with IBA (4000 ppm). Khan *et al*. [20] obtained maximum percentage of bud sprouts from rose cuttings treated with NAA @ 50 mg L^{-1}. They used two auxins viz indole-3-acetic acid (IAA) and naphthalene acetic acid (NAA) at various concentrations and reported NAA superior over IAA in the range of 50 to 75 mg L^{-1}. Jadhav [17] reported the highest percentage of 85.00 of sprouting from stem cuttings of Phalsa treated with NAA @ 200 ppm which was on par with IBA, 200 ppm (81.67%).

Length of the sprouts was also enormously affected by the highest concentration of auxins (NAA). Singh *et al*. [2] observed maximum length of sprouts from February planted stem cuttings of Bougainvillea treated with IBA at the concentration of 3000 mg L^{-1}.

Auxin (NAA) treated cuttings had more potential in increasing rooting percentage, length of the roots and number of roots. Ramadayal *et al*. [21] reported highest percentage of rooting, number of roots per cutting and root length from the hard wood cuttings of Bougainvillea variety Mary Plamer in response to the auxin. Asl *et al*. [22] reported no effect of auxins (IBA) on rooting percentage of Bougainvillea stem cuttings. However they observed maximum number of roots from the cuttings treated with IBA @ 2000 mg L^{-1}. In the present study we recorded much more number of roots with the increasing concentration of NAA up to 6000 mg L^{-1}. The length of roots also increased with the increasing concentration of NAA. The increase in length of roots in cuttings treated with growth regulators may be due to the accumulation of metabolites at the site of application of auxins, cell enlargement, enhanced hydrolysis of carbohydrates, synthesis of new proteins, and cell division induced by the auxins [23]. Contradictory results reported by Asl *et al*. [22]. They reported no significant effect of auxin (IBA) on length of the roots. However they observed maximum root length from control treatment where no auxin was applied. This may be due to the juvenility factor of the plant or different type of auxins used. Junvenility may be the important factor in the rooting potential in different plants [24,25]. Juvenile tissues of woody plants tend to have higher levels of endogenous auxin and are less differentiated and therefore more prone to dedifferentiated [26,10]. Haissig [27] postulated that phenols in juvenile tissues of certain plants tend to be higher than their mature

forms. Also, cuttings were prepared including the apical meristems of stock plants, the region where auxins are synthesized in plants [28].

Gupta *et al*. [29] reported that treatment of Bougainvillea cuttings with 1000 ppm IBA gave maximum rooting (100%) with higher number of roots in soaking method. Panwar *et al*. [30] observed the best rooting in hard wood cuttings of Bougainvillea var. *Alok* treated with IBA 2000 ppm. Jadhav [17] observed more rooting percentage and number of roots from the stem cuttings of phalsa when treated with IBA @ 200 ppm. However he observed more number of leaves from the cuttings treated with NAA @ 200 ppm.

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