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Original Research

The effect of naphthalene acetic acid and some rooting media on rooting abilities and shoot growth of Dwarf Nerium cuttings

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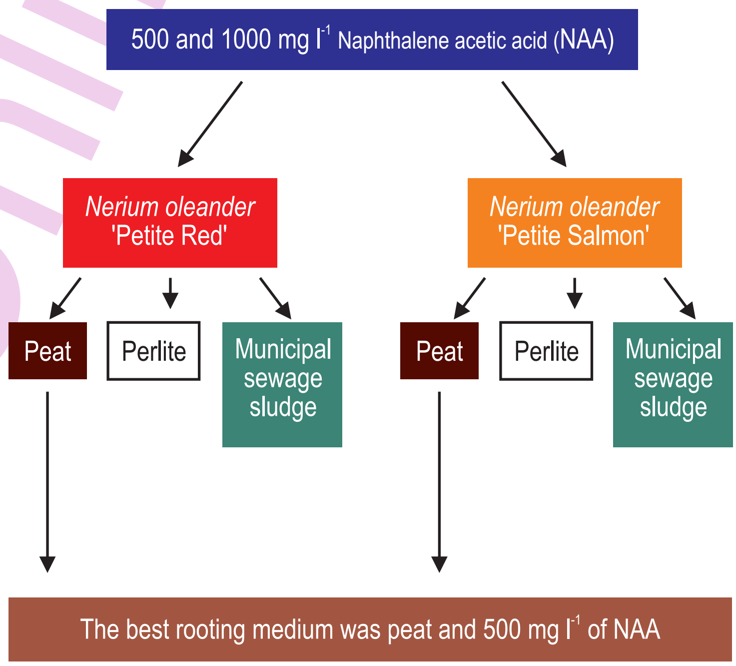
Abstract

Aim: To evaluate the effect of different rooting media and naphthalene acetic acid on rooting abilities and shoot growth of *Nerium oleander* cuttings.

Methodology: Petite Red and Petite Salmon (Dwarf Nerium), the experiment was designed randomly to obtain data from three replicates (ten cuttings per replicate). The cuttings taken during mid February were rooted at different concentrations of NAA (500 and 1000 mg l^{-1}) and in growth media (peat, per lite and municipal sewage sludge) for 45 days. The shoot length and number, fresh and dry weight of shoots, rooting ratios, root number and length, fresh and dry weight of roots were measured.

Results: The results showed that the best rooting medium was peat and 500 mg l^{-1} of NAA increased rooting ratio by 60%, no difference was found between other two NAA concentrations. Moreover, in both varieties, peat growth media plus 500 mg l^{-1} NAA yielded the best results and addition of NAA positively affected shoot fresh and dry weight.

Interpretation: On the basis of this study, it can be concluded that peat was found to be the best rooting media and NAA addition in rooting media positively affected plant growth. Moreover, recycled municipal sewage sludge can be used in ornamental plant cultivation.



Introduction

Nerium (*Nerium oleander* L.), an ornamental plant that belongs to family Apocynaceae is drought resistant, easily grows on seashore, rocky areas and salty soil and can easily adapt to variable conditions. In recent years, dwarf forms obtained as a result of some breeding attempts have become popular as they can be used in many landscape works due to their intensive flowering and compact structures. Moreover, besides being able to adapt to different extreme conditions, they can undergo flowering with different colors for longer periods and can be used for different landscape purposes within indoor plant in pots (Kose and Kostak, 2000). Dwarf Nerium plant is usually preferred in groups for landscape arrangements so that they can induce greater impacts when compared to their solitaire usage.

Dwarf Nerium is a nice ornamental plant with its bushy structure, ever-lasting green color and flowers that can be salmon pink, red, pink and white throughout summer. Production of Dwarf Nerium is usually performed with seed, graft and cutting. Plants obtained through propagation of cuttings not only produce same products characteristically similar to parental plants in addition homogenous products. The research has revealed that the success level in the production of ornamental plants with cutting is affected by internal elements such as genetic structure, plant storage materials and hormones within the plant and external elements such as cutting period, cutting type, humidity, light and bottom heating (Schaberg *et al*., 2000; Ahmed *et al*., 2002; Demirbas, 2010).

Another factor affecting the rooting of Dwarf Nerium cuttings, as in the case of many other ornamental plants, is the application of auxin growth regulator. The research revealed that application of different growth regulators at various concentrations has affected rooting ratios (Swetha, 2005; Uysal *et al*., 2010). Auxins can regulate the growth and development of a plant by affecting many operations including cell prolongation and diversification (Haissig and Davis, 1994; Liu *et al*., 1996; Srivastava, 2002).

Use of appropriate auxin hormone in the production of cutting plays a key role in the rooting of many species as it can increase the amount of carbohydrate carried to the bottom of the cutting and adventitious root formation (Pal, 1995; Gunes, 2000; Smart *et al*., 2003; Rout, 2006). The most widely used growth regulatory substances in the rooting of ornamental plants are Indol Butyric Acid (IBA), Indol Acetic Acid (IAA) and NAA hormones from auxin group (Hartman *et al*., 1997; Kaynak and Ersoy, 1997). These growth regulatory substances are used to produce cuttings with high rates of roots, accelerate rooting and increase the number of roots per cutting (Urgenc, 1982). However, the ratio of rooting in cutting production can vary greatly depending on species and types (Wetswood, 1993; Agaoglu*et al*., 1995). In addition to this, each plant species is rooted with the application of plant growth regulators substances (Kramer and Kozlowski, 1960). Another factor affecting the rooting of cuttings is rooting medium.

In view of the above, the present study was carried out to investigate whether addition of 500 and 1000 mg l^{-1} NAA treatment would enhance the rooting of Dwarf Nerium together with the effect of different growth media on rooting capacities and shoot growth.

Materials and Methods

The experiment was set up in a 150 m^2polyethylene tunnel-type greenhouse in the Ortaca Vocational School of Mugla Sitki Kocman University. As plant materials, 9-13 cm long medial cuttings of Dwarf Nerium varieties of *Neriumo leander* 'Petite Red' (V1: red flowers; V: Variety) and 'Petite Salmon' (V2: salmon colored flowers) by cutting them from their bottom with a slant were used. As the rooting plant hormone, naphthalene acetic acid (NAA) at different concentrations (H1: 0, H2: 500 and H3: 1000 mg l^{-1}) was applied. In the trial, peat (M1), perlite (M2) and municipal sewage sludge (M3) growth media were used and in each medium three replications were carried out and in each

Table 1: Physical and chemical properties of peat material

Parameters	Values
pH	6.44
EC* (dS m^{-1})	1.22
Organic matter (%)	79.10
Total nitrogen (%)	2.23
Available K (mg l^{-1})	2300
Available Na (mg l^{-1})	846
Available Mg (mg l^{-1})	1550

*EC: Electrical conductivity

Table 2: Some physical and chemical properties of municipal sewage sludge

Parameters	Values
pH	7.34
EC* dS m^{-1}	5.0
Organic matter (%)	74.99
Total nitrogen (%)	3.75
Total P (mg l^{-1})	3715
Total K (mg l^{-1})	1081
Total Ni (mg l^{-1})	41.04
Total Cd (mg l^{-1})	0.77
Total Al (mg l^{-1})	2575
Total Fe (mg l^{-1})	5252
Total Pb (mg l^{-1})	9.33
Total Cu (mg l^{-1})	15.81
C/N ratio	11.60

*EC: Electrical conductivity

replication 10 cuttings; thus, a total of 540 cuttings were selected according to random parcel trial design. Due to this system, each pot was placed randomly in the experiment and turned around itself for taking environmental factors equally.

Medium-size perlite having pH value of 6.5 and peat and municipal sewage sludge (some of whose physical and chemical properties are presented in Table 1 and 2) were used in the trial. In the peat and municipal sewage sludge samples, organic matter was determined by the method of Nelson and Sommers (1982), total nitrogen as per Bremmer (1965), P, K, Ca, Na and Mg according to Kacar (1995) and Fe, Zn, Mn and Cu analyses according to Lindsay and Norvell (1978), respectively.

Throughout the trial period, the average temperature was calculated to be 14.7 °C with minimum 10.7 °C and maximum 22.1 °C and relative humidity was found to be around 74.9%. The cuttings were taken into rooting tables for rooting without subsurface soil heating. Some calculations were done to determine the effect of different amounts of hormone treatments and the rooting media on the ratios of the cuttings rooted and their rooting and development. The trial lasted throughout the 45-day rooting period. For the purpose of rooting, NAA hormone was applied to the bottoms of the cuttings through quick dipping method for 5 sec. At the end of the trial period, the cuttings were removed and then the values related to criteria such as number of shoots on the cuttings, shoot length (cm),shoot fresh and dry weight (g), rooting ratio (%), the number of existing roots (per cutting), root length (cm), root fresh and dry weight (g) were determined. The data collected throughout the trial were analyzed through SPSS 15.0 package program and variance analysis was carried out.

Results and Discussion

The highest rooting ratio and root length were found in V2 (salmon color), whereas M2 (Perlite) was observed to be potential supporter of rooting ratio and root length in comparison other

Table 3: Rooting ratio, root number and root length of cuttings

Treatments	Rooting ratio (%)	Root number (number per cutting)	Root lenght (cm)
V1	25.67^{b}	5.22^{b}	2.42^{b}
V2	58.63^{a}	5.96^{a}	4.04^{a}
LSD	4.801**	0.493**	0.510**
M1	59.06^{a}	8.50^{a}	2.81^{b}
M2	53.56^{a}	6.67^{b}	5.28^{a}
M3	13.83^{b}	1.61^{c}	1.60^{c}
LSD	5.88**	0.604**	0.624**
H1	29.00^{b}	4.50^{c}	3.29
H2	47.22^{a}	6.94^{a}	3.32
H3	50.22^{a}	5.33^{b}	3.08
LSD	5.88**	0.604**	ns
V1xM1xH1	7.00^{d}	4.00^{f}	1.50
V1xM1xH2	47.00^{c}	12.00^{b}	2.46
V1xM1xH3	47.00^{c}	8.00^{c}	1.04
V1xM2xH1	20.00^{d}	7.00^{cd}	4.93
V1xM2xH2	40.00^{c}	7.00^{cd}	3.48
V1xM2xH3	40.00^{c}	5.00^{ef}	4.24
V1xM3xH1	7.00^{d}	2.00^{g}	2.00
V1xM3xH2	10.00^{d}	1.00^{g}	1.50
V1xM3xH3	13.00^{d}	1.00^{g}	0.63
V2xM1xH1	73.00^{b}	5.00^{ef}	3.50
V2xM1xH2	93.33^{a}	14.00^{a}	4.23
V2xM1xH3	87.00^{ab}	8.00^{c}	4.12
V2xM2xH1	47.00^{c}	7.00^{cd}	6.05
V2xM2xH2	80.00^{ab}	6.00^{de}	6.26
V2xM2xH3	94.33^{a}	8.00^{c}	6.73
V2xM3xH1	20.00^{d}	2.00^{g}	1.73
V2xM3xH2	13.00^{d}	1.67^{g}	2.00
V2xM3xH3	20.00^{d}	2.00^{g}	1.73
LSD	14.40*	1.48**	ns

V1: Red, V2: Salmon, M1: Peat, M2: Perlite, M3: Sewage sludge, H1: 0 mg l^{-1}, H2: 500 mg l^{-1}, H3:1000 mg l^{-1} V: Variety, M: Growth media, H: Plant hormone; ns: non significant; Withineachcolumn, values followed by same letters are not significantly different according to LSD test, ** Significant at p<0.01, * Significant at p<0.05 level

Table 4 : Data about the shoot number of cuttings and shoot length

Treatments	Shoot number (number per cutting)	Shoot length (cm)
V1	4.44^{a}	5.47^{b}
V2	3.67^{b}	7.34^{a}
LSD	0.552**	0.552**
M1	4.50	7.19^{a}
M2	3.67	7.45^{a}
M3	4.00	4.58^{b}
LSD	ns	0.667**
H1	3.67	5.50^{b}
H2	4.50	6.64^{a}
H3	4.00	7.08^{a}
LSD	ns	0.667**
V1xM1xH1	3.00^{cd}	3.50^{de}
V1xM1xH2	6.00^{a}	7.60^{ab}
V1xM1xH3	5.00^{ab}	7.60^{ab}
V1xM2xH1	4.00^{bc}	5.20^{cd}
V1xM2xH2	3.00^{cd}	7.94^{ab}
V1xM2xH3	4.00^{bc}	8.30^{a}
V1xM3xH1	6.00^{a}	4.80^{cd}
V1xM3xH2	5.00^{ab}	2.30^{f}
V1xM3xH3	4.00^{bc}	2.00^{f}
V2xM1xH1	4.00^{bc}	8.20^{a}
V2xM1xH2	5.00^{ab}	8.11^{a}
V2xM1xH3	4.00^{bc}	8.11^{a}
V2xM2xH1	3.00^{cd}	7.29^{ab}
V2xM2xH2	4.00^{bc}	7.49^{ab}
V2xM2xH3	4.00^{bc}	8.46^{a}
V2xM3xH1	2.00^{d}	4.00^{de}
V2xM3xH2	4.00^{bc}	6.40^{bc}
V2xM3xH3	3.00^{cd}	8.00^{a}
LSD	1.66*	1.66**

V1: Red, V2: Salmon, M1: Peat, M2: Perlite, M3: SewageSludge, H1: 0 mg l^{-1}, H2: 500 mg l^{-1}, H3:1000 mg l^{-1} V: Variety, M: Growth Media, H: PlantHormone; ns: nonsignificant; Within each column, values followed by same letters are not significantly different according to LSD test, ** Significant at $p<0.01$, * Significant at $p<0.05$ level

media. The effect of NAA treatment on rooting and root length is quite positive in that when compared to the control, 500 mg l^{-1} NAA treatment increased rooting ratio by 60%. The effects of variety, medium and NAA combinations were separately determined, and thus the best effect in V1 and V2 was obtained in M1xH2 combination (Table 3).

When the data collected in relation to the number of shoots in the cuttings were examined, it was found that while the number of shoots was higher in V1, more positive results were given by V2 in terms of shoot length. When the media were evaluated, the shoot length was found to be higher in M1 and M2 than that of M3. When comparative analyses were done most optimal number and length of shoots were obtained in both V1 and V2 with M1xH2 combination (Table 4).

When the shoot and root fresh and dry weights of the cuttings were determined, it was found that V1 had more dry matter content than V2. When the media were evaluated, it was found that M1 andM2 media were more advantageous than M3. Another finding of the study was that the addition of NAA into a medium considerably increased shoot fresh and dry weight. Among the combinations, V1xM1xH2 and V2xM2xH2 combinations were found to have yielded the best results (Table 5).

Auxins plant growth hormones effectively contribute in cell division of plants and accelerate growth and development, can act in multiple directional and polar manner and can be transferred from cell to cell. NAA, as an effective rooting hormone, is widely used commercially for this purpose (Kumlay and Eryigit, 2011). Commercial and scientific applications indicate that the use of NAA, IAA and IBA varieties of auxin is widespread. Uysal *et al*. (2010) attempted to root *Rosmarinus officinalis* cuttings with three different NAA doses (100, 500 and 1000 mg l^{-1}) and the highest number of roots (n=31) was obtained as a result of 1000 mg l^{-1} NAA treatment. Lavender (*Lavundula officinalis)*, which is a

Table 5: Fresh and dry weights of shoot and root cuttings

Treatments	Root fresh weight (g)	Root dry weight (g)	Shoot fresh weight (g)	Shoot dry weight (g)
V1	0.30^{a}	0.06^{a}	2.42^{a}	0.50^{a}
V2	0.18^{b}	0.04^{b}	1.60^{b}	0.41^{b}
LSD	0.046**	0.05**	0.050**	0.053**
M1	0.34^{a}	0.07^{a}	2.31^{b}	0.54^{a}
M2	0.33^{a}	0.06^{b}	2.38^{a}	0.55^{a}
M3	0.04^{b}	0.01	1.33^{c}	0.28^{b}
LSD	0.056**	0.006**	0.062**	0.064**
H1	0.21^{b}	0.04^{b}	1.99^{b}	0.48
H2	0.28^{a}	0.05^{a}	2.15^{a}	0.43
H3	0.22^{b}	0.04^{b}	1.88^{c}	0.46
LSD	0.056*	0.006**	0.062**	ns
V1xM1xH1	0.39	0.095^{a}	3.39^{a}	0.66^{b}
V1xM1xH2	0.61	0.095^{a}	2.04^{e}	0.45^{d}
V1xM1xH3	0.56	0.085^{ab}	2.65^{c}	0.55^{b}
V1xM2xH1	0.30	0.060^{de}	3.35^{a}	0.71^{ab}
V1xM2xH2	0.37	0.065^{c}	1.83^{f}	0.40^{d}
V1xM2xH3	0.40	0.075^{bc}	2.16^{e}	0.47^{d}
V1xM3xH1	0.02	0.013^{fg}	1.20^{hj}	0.21^{e}
V1xM3xH2	0.01	0.008^{fg}	3.41^{a}	0.67^{b}
V1xM3xH3	0.01	0.008^{fg}	1.70^{fg}	0.40^{d}
V2xM1xH1	0.15	0.055^{e}	1.60^{g}	0.57^{b}
V2xM1xH2	0.23	0.070^{cd}	2.44^{d}	0.61^{bc}
V2xM1xH3	0.13	0.035^{f}	1.71^{fg}	0.44^{d}
V2xM2xH1	0.33	0.013^{fg}	1.34^{h}	0.49^{c}
V2xM2xH2	0.41	0.060^{de}	2.67^{c}	0.36^{d}
V2xM2xH3	0.17	0.055^{e}	2.94^{b}	0.87^{a}
V2xM3xH1	0.08	0.013^{fg}	1.07^{g}	0.23^{e}
V2xM3xH2	0.07	0.009^{fg}	0.51^{k}	0.12^{e}
V2xM3xH3	0.05	0.008^{g}	0.085^{l}	0.02^{f}
LSD	ns	0.014*	0.151**	0.158**

V1: Red, V2: Salmon, M1: Peat, M2: Perlite, M3: SewageSludge, H1: 0 mg l^{-1}, H2: 500 mg l^{-1}, H3:1000 mg l^{-1} V: Variety, M: Growth Media, H: PlantHormone; ns: nonsignificant; Within each column, values followed by sameletters are not significantly different according to the LSD test, ** Significant at $p<0.01$, * Significant at $p<0.05$ level

half-bushy and long-lasting plant, was treated with NAA and IBA (1000, 2000 and 3000 mg l^{-1}) for rooting and the highest rooting ratio (86%), the highest number of roots (700), root length (10.3 cm) and survival ratio in the field (93%) were obtained as a result of 3000 mg l^{-1} IBA treatment (Bhat *et al*., 2008).

Nerium oleander cuttings with low starch and sugar content were treated with 2000 mg l^{-1} IBA, they exhibited better rooting. In their study focused on the reproduction of Indian lavender with cutting stated that rooting medium, type of cutting, the season in which it is taken, preliminary applications and environmental conditions affect the rooting success of cutting.

Cekic *et al*. (2013) investigated the effect of IBA on medial cutting of mulberry plant. With 6000 mg l^{-1} IBA treatment, 40% rooting was obtained in purple mulberry plant. And again when IBA was applied, the highest number of roots per cutting was attained.

In an another study, the effects of cutting period, rooting medium and IBA (indole 3 butyric acid) on the rooting of *N. oleander* L. cv variegata were investigated. IBA treatments conducted as 5-second tip layering (4000, 6000 and 8000 mg l^{-1}) positively affected rooting, the number of roots and root length; yet, the highest rooting ratio (47.7%) was obtained in the cuttings treated with 1000 mg l^{-1} IBA application (Kose and Kostak, 2000).

Similar studies were conducted on Nerium, where 19% rooting was attained in half-medial cuttings taken in July and treated with 8000 mg l^{-1} IBA, 70% rooting was achieved in the cuttings taken in February. These results show that cutting period affects rooting. In the current study, 6000 mg l^{-1} IBA was found to have adverse effects on rooting.

In a study investigating the effect of hormone doses on cutting rooting of *Lavandula stoechas*, it was found that hormone doses positively affected rooting and IBA applied to cuttings increased rooting ratio, root length and root number. The highest rooting ratio (70%) was obtained from the cuttings treated with 4 000 mg l^{-1} IBA (Ayanoglu *et al*., 2000).

Municipal sewage sludge having a potential of being used in agriculture, forests, parks and gardens and grass areas and in recent years has also has been widely used in land recreation, urban landscape design and arboriculture (Yalcin *et al*., 2011). When used in the above-mentioned areas, municipal sewage sludge can be both disposed and constitute a source of fertilizer. Within the current study, when compared to other media, municipal sewage sludge was found to be the weakest medium. In a study where municipal sewage sludge was used, municipal sewage sludge applied to the soil resulted in slight improvement in plant growth and when it was repeatedly applied or applied at high doses, phytotoxicity and high heavy metal content were detected (Topcuoglu *et al*., 2003). Moreover, municipal sewage sludge added to the growth media in *Limonium sinuatum* cultivation was found to have some important effects on the number of flowers, root length, pedicle length, number of leaves and plant upper side fresh weight (Akat*et al*., 2015). In another study, Suchkova *et al.* (2014), investigated the macro and micro element contents in *Cyperus rotundus, Lolium perenne* and *Lycopersicon esculentum* species cultivated in municipal sewage sludge medium and reported that the macro element uptake of plants increased and this affected positively plant growth. Xue and Huang (2013) in their study called the Effect of Municipal Sewage Sludge Compost on the Growth of Common Peony found that soil microbial mass, basal respiration, soil organic carbon ratio, enzyme activities and plant height, pedicle and number of flowers per plant were positively affected by municipal sewage sludge treatment. As the nutritional value of municipal sewage sludge vary depending on its constituents, it may have some how limited effects on plant growth. Peat derived from peat (moss) litters is a plant growth media widely used in seedling cultivation of ornamental plant sand vegetables.

In a study conducted to investigate the cultivation of cucumber seedling in different organic and inorganic media, as the growth media, pulp residue, perlite, slag and peat were used and the best outcomes were obtained in terms of seedling height, weight and root length in the mixture of peat and perlite (Cinkilic, 2008).

In a study investigating the development of *Rosmarinus officinalis*, *Euonymus japonicas* and *Salvia officinalis* cuttings, three different rooting media peat, soil and sand were used. In terms of ratio of rooted cutting, the best rooting medium was found to be soil; however, in peat medium, both the root development and plant upper side development were found to be better (Celik *et al*., 2010).In another study, as media, slag, perlite and peat were used and the root length of the plants growing in peat medium was found to be higher than the others growing in the other media. When the development of plants was evaluated, the best results were obtained in peat medium (Hepaksoy and Tanrisever, 2004).In the current study, peat was also found to be the best rooting medium.

Findings of the present study suggest that NAA contributed to rooting and plant growth of Dwarf Nerium plant cuttings. While peat was found to be the best rooting medium, NAA addition was observed to have positively affected rooting ratio and general development. Further, study also suggest that municipal sewage sludge as plant growth media could be used after being subjected to recycling in commercial ornamental plant cultivation.

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