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Propagation of a rare medicinal plant species *Premna integrifolia* by hardwood cuttings

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ABSTRACT

An investigation was carried out to study the effect of different growth regulators on rooting of *Premna integrifolia* cuttings, a rare medicinal plant species. The various root and shoot parameters recorded significantly higher values when the cuttings were treated with growth regulators as compared to control. The higher rooting (86%) was recorded in the cuttings treated with 1000 ppm IBA against control (67.66%). The other parameters like number of roots per cutting, fresh weight and dry weight of roots, sprouting percentage, field establishment percentage *etc.* were also recorded higher in the cuttings treated with IBA 1000 ppm (96.5%) followed by IBA 2000 ppm (93.20%).

Key words: *Premna integrifolia*, propagation, indole butyric acid, stem cuttings.

INTRODUCTION

Premna integrifolia Linn. (syn. *Premna serratifolia* Linn.) is commonly known as *Agnimantha, Jayantee, Shreeparna*, *munna*, *munnai*, that belongs to the family Verbanaceae. It is a large shrub or a small tree upto nine metres in height with yellow bark, brown woody aromatic root. Leaves are simple, opposite, ovate, membranous and irregularly toothed. Flowers are small greenish-white with strong disagreeable odour. Fruits are glabrous, black when ripened and saucer shaped calyx surrounding the base (Deshapande, 4). Almost all parts of this plant, *i.e.* root, leaf and bark have numerous medicinal uses. The roots are astringent, anti-inflammatory, cardio-tonic, digestive, stomachic, carminative, antibacterial and tonic (Sivarajan and Indira, 19). The roots are used in many *Ayurvedic* preparations and it is one of the main ingredients in *Dashamula*. As reported by Chopra *et al.* (2), the juice from the bark is used as a powerful anti-malarial agent. It is also reported that the stem bark contains alkaloid premnin, which decrease forces of contraction of hearts. It also has a role in dilation of pupils.

Premna integrifolia is classified as a rare medicinal plant with a lot of medicinal value. It is also considered as one of the important folk medicines used since ancient times (Khare, 9). Besides its medicinal use, it is also used for fodder purpose. Though it can be multiplied through seeds, continuous destruction of the trees for fodder purpose is also a matter of concern. The present investigation aims at fulfilling the above needs by attempting its asexual propagation using hardwood cuttings, besides conservation of this species.

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MATERIALS AND METHODS

The experiment was carried out during 2009 between the months of January-May. The experimental site was located at Medicinal and Aromatic Plants Unit, Saidapur farm, Department of Horticulture, University of Agricultural Sciences, Dharwad. The hardwood cuttings were procured from the coastal region of Gokarn, Uttara Kannada district, Karnataka. The median portion of the stem was used for propagation of cuttings with a length of 15 cm without any branches and leaves, leaving 4-5 buds per cutting with uniform thickness (25 mm dia.). The basal portion of the cutting was given a slant cut and lower bud of the cutting was planted in the medium. Finely powdered sieved farm yard manure, sand and peat in the ratio of 1:2:1 was taken and filled into 3' × 5' sized perforated polythene bag of 200 micron thickness. Before planting, media was drenched with carbendazim (0.2%) as a prophylactic measure against fungal diseases. The experiment was laid out in completely randomized design with three replications and 25 cuttings per replication. Indolebuteric acid and coumarin at 500, 1000, 2000 ppm were taken as growth regulator treatments. For treating the cuttings with growth regulators, the cuttings were dipped 1.5 to 2.00 cm deep in the growth regulator solution for two min. and planted into polythene bags. The experiment was carried out in the polytunnel (with three feet height and width) under partial shade condition (50% shade). This structure helped for maintaining optimum temperature and higher and constant relative humidity (an average temperature of 29°C; relative humidity of 75% and light intensity of 3500 lux was recorded inside the polytunnel during the period of experimentation), which in turn increased the rooting percentage in the cuttings. The polybag raised cuttings were watered regularly. After three months, the rooted cuttings were carefully taken out of the polythene bag without damaging the roots (five in number from each replication and each treatment) and were washed in water for some time to remove the soil. Observations in relation to roots such as number of roots per cutting, length of longest root, fresh and dry weight of roots were taken. Other observations included were bud sprout percentage, number of leaves per cutting, number of sprouts per cutting and per cent rooting. The data was analysed statistically as per the method suggested by Panse and Sukhatme (12).

RESULTS AND DISCUSSION

The data pertaining to various root and growth parameters as influenced by different growth regulators are presented in Tables 1 and 2, respectively. The growth regulator treatments had a significant effect on different root and shoot parameters. There was a significant influence of treatments on rooting parameters such as per cent rooting, number of roots per cutting, length of the longest root, fresh weight and dry weight of roots. Higher rooting (86%) was registered in the cuttings treated with 1000 ppm IBA followed by 2000 ppm IBA (81.24%) which were significantly superior over control. This may be due to the action of IBA, which might have caused hydrolysis and translocation of carbohydrates and nitrogenous substances at the base of cuttings and resulted in accelerated cell elongation and cell division in suitable environment (Hartmann *et al.*, 7). Earlier, Shwetha (18) reported better induction of rooting (66.6%) in Indian lavender cuttings treated with 2000 ppm IBA as compared to control (15.3%).

Among different growth regulator treatments, the number of roots per cutting was significantly higher in the cuttings treated with 1000 ppm IBA (42) followed by IBA 2000 ppm (37.13). Increased number of roots due to auxin application is a common feature in many herbaceous perennial crops (Haissing and Davis, 6; Hartmann *et al.*, 7). Similar results were obtained by Sandhu *et al.* (16) while working with pear rootstock Kaith stem cuttings. They obtained higher number of roots per cutting, rooting percentage and length of roots in cuttings treated with 200 ppm IBA as compared to control. Higher number of roots in cuttings treated with IBA may be ascribed to the higher partitioning of photosynthates towards root development.

Maximum length of the root was recorded in the cuttings treated with IBA 1000 ppm (13.50 cm) followed by IBA 500 ppm (12.20 cm). The effect may be due to slow translocation property or slow destruction by auxin destroying enzyme system (Debnath and Maiti, 3). The fresh and dry weight of roots recorded higher values in the cuttings treated with IBA 1000 ppm (393 and 91 mg, respectively), which were statistically superior to control (85 and 28 mg, respectively). The higher dry weight of the roots may be attributed to increased number of roots and length of longest root. Similar

Table 1. Rooting percentage, number of roots per cutting, root length, fresh weight and dry weight of roots as influenced by different growth regulator treatments in *P. integrifolia* stem cuttings.

Treatment	Rooting (%)	No. of roots per cutting	Length of longest root (cm)
Control	67.66 (55.32)*	25.00	6.93
IBA 500 ppm	76.20 (60.78)	34.70	12.20
IBA 1000 ppm	86.00 (68.00)	42.00	13.50
IBA 2000 ppm	81.24 (64.31)	37.13	11.60
Coumarin 500 ppm	79.00 (62.70)	26.13	10.20
Coumarin 1000 ppm	63.00 (52.51)	25.77	10.00
Coumarin 2000 ppm	61.66 (51.72)	25.33	10.10
CD at 5%	2.59	4.61	1.28

*Figures in parentheses indicate Arc sine transformed values.

Table 2. Sprouting percentage, number of sprouts and number of leaves per cutting, Shoot length and field establishment per cent as influenced by different growth regulator treatments in *P. integrifolia* stem cuttings.

Treatment	Sprouting (%)	No. of sprouts per cutting	No. of leaves per cutting	Shoot length (cm)	Field establishment (%)
Control	73.00 (58.67)*	3.06	29.00	15.20	86.20 (68.17)*
IBA 500 ppm	81.00 (64.13)	3.73	38.46	30.90	92.50 (74.08)
IBA 1000 ppm	88.30 (69.93)	4.00	40.10	33.60	96.50 (79.19)
IBA 2000 ppm	86.66 (68.55)	4.26	31.00	26.60	93.20 (74.85)
Coumarin 500 ppm	81.70 (64.65)	3.80	30.93	23.40	90.00 (71.54)
Coumarin 1000 ppm	71.69 (57.83)	4.26	28.20	17.66	84.00 (66.40)
Coumarin 2000 ppm	66.66 (54.71)	4.80	32.53	16.00	82.40 (65.17)
CD at 5%	1.68	0.67	5.05	4.19	3.56

*Figures in parentheses indicate Arc sine transformed values.

effect has also been observed by Farooqi *et al.* (5) in *Rosa damascena.*

Shoot parameters like sprouting percentage, number of sprouts, number of leaves and shoot length was also found to be maximum in cuttings treated with IBA 1000 ppm. The maximum sprouting percentage was observed in IBA 1000 ppm (88.30) followed by IBA 2000 ppm (86.66), which were highly significant to all the treatments including control (73.00). Srivastava *et al*. (20) reported maximum rooting and establishment percentage (87.66%) in *Kiwi* plant (*Actinidia chinensis*) cuttings treated with IBA at 5000 ppm against control (12%). They also obtained maximum number of roots per cutting (52.5) and length of the longest root (21.53 cm) compared to control (15 and 6.5 cm, respectively). Similar results were obtained in *Jatropha curcas* and *Pongamia pinnata* cuttings treated with IBA by Noor *et al*. (11), and Vigya *et al*. (21), respectively. The data on number of sprouts per cutting was not significantly influenced by different growth regulator treatments. However, the higher number of sprouts was recorded in the cuttings treated with IBA 2000 ppm (4.26) followed by the cuttings treated with IBA 2000 ppm (4.00). This may be due to increased auxin concentration which increased rooting and enhanced sprouting in cuttings. Different concentrations of IBA caused significant difference over control with regard to number of leaves developed per cutting. The higher number of leaves was registered in the cuttings treated with IBA 1000 ppm (40.10) followed by the cuttings treated with IBA 500 ppm (38.46). Wiesman and Shimon (22) had also observed similar results in olive cuttings treated with IBA. The possible reason for such increase may be due to the activation of shoot growth which probably increased the number of nodes that lead to development of higher number of leaves.

Maximum shoot length (33.60 cm) was recorded in IBA 1000 ppm against control (15.20 cm). The probable reason for increase in length of shoot may be the better utilization of carbohydrates, nitrogen and other nutrients, which was aided by growth regulators (Chandramouli, 1). The increase in the length of the shoot with the increase in concentration of IBA was also noted by Shivanna *et al.* (17) in *Jeevanthi* (*Leptadenia reticulata*). Number of internodes was recorded maximum in IBA 2000 ppm and coumarin 1000 ppm treatment (Fig. 1), however there is no significant difference between the number and length of internodes among the different treatments.

Total fresh biomass and dry matter content were found to be significant in IBA 1000 ppm over control which is depicted in Fig. 2. The higher field establishment percentage (96.50) was noticed in the cuttings treated with IBA 1000 ppm followed by IBA 2000 ppm (93.20). Legesse (10) recorded similar observations while propagating African pencil wood cedar, an endangered species. Better shoot parameters in turn enhanced the rooting in cuttings and as the IBA

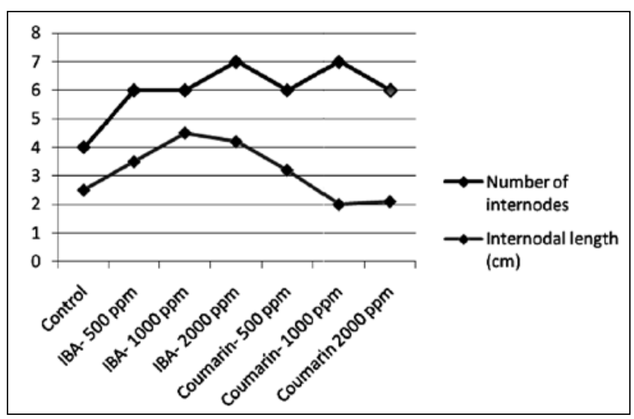

Fig. 1. Effect of different growth regulators on number of internodes and internodal length (cm) of *P. integrifolia* cuttings.

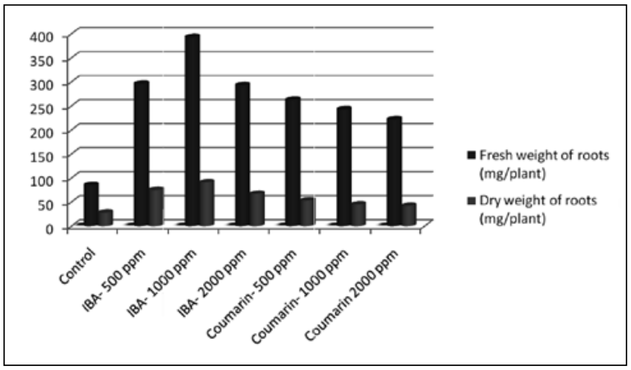

Fig. 2. Effect of different growth regulators on fresh and dry root biomass content (mg/plant) in *P. integrifolia.*

concentration increased, rooting and sprouting per cent also increased. Similar results were obtained in *Rauvolfia* cuttings treated with IBA 2000 ppm (Husen, 8) and there was significant increase in rooting and sprouting percentage against the control.

The root and shoot parameters were slightly inhibited in the cuttings treated with higher concentration of IBA at 2000 ppm. Application of synthetic auxins at high concentration can inhibit bud and root development. Under such circumstances, IBA was translocated to upper part of the cutting, where it inhibited bud growth and increased ethylene synthesis of the cuttings (Hartmann *et al*., 7). Similar results were obtained in *Annona squamosa* cuttings treated with IBA wherein the root parameter values were higher in cuttings treated with IBA 5000 ppm and IBA 10000 ppm had clearly inhibited rooting (Praveen, 13).

Number of sprouts per cutting was recorded significantly higher in coumarin 2000 ppm than all other treatments. Similar results were obtained in *Oxytenathera stocksii* cuttings treated with 10 ppm coumarin (highest concentration tried) showed higher sprouting intensity, biomass production and seedling establishment as compared to the rest of the treatments (Reddy, 15). Ramayanake *et al*. (14) had also got better rooting in three species of bamboo nodal cuttings treated with coumarin. In *P. integrifolia*, coumarin at 500 ppm promoted both shoots and root growth but at higher concentrations it inhibited the growth. Although coumarin is a growth retardant, at lower concentration it promotes rooting and shooting. However, it induced abnormal shoot growth and dwarfness in *P. integrifolia* at 2000 ppm.

Among the different treatments, IBA at 1000 ppm was found to be the best, which recorded maximum root and shoot parameter values. The cuttings treated with IBA at 2000 and 500 ppm also recorded better root parameters next to IBA 1000 ppm. As the rooting and field establishment was better with IBA 1000 ppm, which may helps in large scale multiplication of the species. It also helps for cultivation and supply of raw material to pharmaceutical industries. The results of the present investigation may also be helpful for the institutions, those working on biodiversity conservation.

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