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Clonal propagation of *Dalbergia sissoo* through stem cuttings

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Abstract
Vegetative propagation plays a key role in tree improvement programmes as a tool for large scale multiplication of superior genotypes. *Dalbergia sissoo* is a globally important tree species and it is used for timber purposes. Multiplication of clones through vegetative propagation is very difficult in *Dalbergia sissoo*. To overcome, different concentrations of IBA solutions *viz*., 1000 ppm (T1), 2000 ppm (T2), 3000 ppm (T3) 4000 ppm (T4), 5000 ppm (T5) were prepared and the stem cuttings prepared from the mother tree were dipped in them. Untreated cuttings served as control (T6). The results of the present showed, cuttings of the size of 15 cm X 2.5 cm *Dalbergia sissoo* dipped with IBA 4000 ppm planted in Sand: Soil: VAM (2:1:1) is suitable for enhancing rooting and survival percent of stem cuttings. Such knowledge will aid propagators, geneticists and tree improvement specialists in selecting treatments that reduce propagation costs by maximizing rooting success. Hopefully the present studies will also pave the way to develop clones of superior trees of *Dalbergia sissoo* used for nitrogen fixation, fodder in agroforestry or large scale timber purposes.

Keywords: Clonal propagation, *Dalbergia sissoo*, plant growth harmones

Introduction
Vegetative propagation plays a key role in tree improvement programmes as a tool for large scale multiplication of superior genotypes. *Dalbergia sissoo* is a globally important tree species and it is used for timber purposes. This species occurs through out the sub-Himalayan tract and outer Himalayan valleys from the Indus to Assam. Usually upto 900 m, but occasionally ascending to 1500 m. The tree has also been introduced in the plains of West Pakistan through canal irrigation. The tree has been introduced in Java and Nigeria, Mauritius, Srilanka, Kenya, Northern Palestine, Rhodesia and South Africa with varying degrees of success. According (Troup, 1921) [14], it is likely that sissoo is indigenous only to the sub-Himalayan and Bhabaar areas and has been introduced by man elsewhere. Shisam grows in subtropical to tropical climate. In its natural habitat, the absolute maximum shade temperature varies from about 39 C to 49 C and the absolute minimum shade temperature from about 4-6 C. The average annual rainfall varies from 760-4570 mm bulk of which is received in 3-4 months precedes and followed by a long dry season. The tree can be grown on almost any site including pure stand, provided the drainage is good and sufficient soil-aeration is available. The tree has a marked preference for porous soil with adequate moisture. It comes up on new sandy alluvial well drained soils. The tree is a strong light demander right from the seedling stage being a pioneer tree species; it requires full overhead light for successful regeneration and establishment. The seedlings are comparatively sensitive to drought. Shisham is a moisture loving species but it cannot tolerate bad soil drainage. It responds to irrigation very well. Naturally regenerated sissoo trees are usually wind-firm due to their well developed root systems but wind falls are not uncommon in irrigated plantations. Shisham has a deep tap root and long lateral roots of considerable length and thickness. Root success sprout up from the lateral roots when they are exposed or cut.

Vegetative propagation
Vegetative propagation is the potential means of raising *Dalbergia sissoo* plantations inorder to avoid the dieback disease and other associated problems like forking in tree trunk, poor growth and lack of clean bole etc. For many economically important tree species vegetative propagation techniques have already been used for maintaining genetic superiority and

increasing productivity (Tewari, 1994) [13]. Further, planting materials of asexual reproduction can be raised almost throughout the year in shorter time (James, 2000; Kumar 2010) [3, 5]. However standardization of vegetative propagation techniques at species level is essentially required for mass multiplication of planting stock. Sissoo can be vegetatively propagated by root stem cuttings, root cutting or by root suckers. (Nanda, 1970) [9]. Stem cuttings of sissoo roots easily and the rooting response varies with season. Age and mother of the vigour plant do not affect rooting potential of stem cutting, but site conditions from where cuttings are taken, may affect it. IAA has generally been reported to be more effective than the other auxins, in promoting rooting during summer months (Nanda, 1970) [9]. While IBA and Indole Propionic Acid were found effective during winter (Lush and Jaya, 1940) [7]. Rootings from cuttings obtained from 4 years and 30 years old shisham trees and treated with different concentrations of IAA, IBA and NAA has showed that in the February planted cuttings of younger trees, IBA 100 ppm produced maximum rooting (50%) followed by IAA 100 ppm (40%). In case of cuttings of old trees, the IBA 100 ppm gave 30% rooting. It is one of the few NFT's and timber is strong and very elastic. The wood is suitable for handles for striking tools, scooping, cutting and shaping tools. Sissoo is eminently suitable for railway sleepers, but due to its great usefulness in constructional and cabinet purposes, it is rarely used for railway sleepers. It is a preferred species for building railway coaches, truck and lorry bodies. It is one of the best timbers for turnery works and can be worked into intricate, deep and delicate ornamental carvings. Vegetative clonal propagation is generally considered as an important tool for true to type of planting stock with the highest genetic quality materials. (Nanda1970) [9]. Exogenous auxins are commonly used to induce rooting efficiency and quality if stem cutting in most of the forest tree species. Inorder to reduce plant variability and at the same time ensured increased productivity the development of vegetative/ clonal propagation techniques is required. For the production of high quality timber and faster tree growth it is essential to start by selecting superior clones from which the stem shoot cuttings are to be taken. However well documented scientific study in this species is scant with regard to the efficient hormonal treatment and exact concentration. The objective of the present study is to evaluate the effect of different length and diameter of the cuttings, different rooting media and identifying optimum concentration of IBA.

Materials and Methods

The experiment was conducted at Forest College and Research Institute, Mettupalayam located at 11 9 N latitude and 77 56 E Longitude at an altitude of 300 m above MSL. The climate is semi-arid tropical type with hot summer and cold-winter. The dry season starts from early February to mid June and wet season from mid August to early November. The mean annual rainfall in the region is 895 mm distributed over 49 rainy days with north east monsoon contributing to 60% and the balance through summer showers and southwest monsoon. The mean maximum and minimum temperature are 30 c and 20 c respectively. The experimental materials selected for the studies consisted of stem cuttings from plus trees of *Dalbergia sissoo*, and the rooting study was conducted in lowcost polytunnels. This experiment was laidout in a completely randomized block design with 3 replications.

Standardization of concentration of IBA

Different concentrations of IBA solutions *viz*., 1000 ppm (T1), 2000 ppm (T2), 3000 ppm (T3) 4000 ppm(T4), 5000 ppm (T5) were prepared and the stem cuttings prepared from the mother tree were dipped in them. Untreated cuttings served as control (T6). The treated cuttings along with control were then planted in polybags filled with rooting media and kept in the low cost poly tunnels for rooting.

Table 1: Optimization of IBA concentration for clonal propagation of *Dalbergia sissoo*

Sl. No	Treatments	Rooting %	Root number per cutting	Root length per cutting	Survival %
1	T1-1000 ppm	60.00	22.00	9.52	60.00
2	T2-2000 ppm	80.00	32.66	8.10	76.60
3	T3-3000 ppm	70.00	34.00	8.95	60.00
4	T4-4000 ppm	86.60	56.66	15.77	76.60
5	T5-5000 ppm	70.00	27.33	9.15	60.00
6	T6-Control	23.30	4.00	4.33	23.30
	Grand Mean	64.16	29.44	9.33	64.98
	S.Ed	0.63	0.44	0.97	0.54
	C.D (0.05)	1.40	0.95	2.11	1.18

Table 2: Effect of size of the stem cuttings on clonal propagation of *Dalbergia sissoo*

Sl. No	Treatments	Rooting %	Number of primary roots	Root length per cutting	Survival %
1.	T1-10 cm X 1.0 cm	50.00	5.70	5.19	40.00
2.	T2-10 cm X 1.5 cm	53.33	6.10	6.29	43.30
3.	T3-10 cm X 2.0 cm	56.66	6.26	6.15	50.00
4.	T4-15 cm X 1.0 cm	66.66	6.23	5.70	50.00
5.	T5-15 cm X 1.5 cm	56.67	5.50	5.46	50.00
6.	T6-15 cm X 2.0 cm	75.80	16.1	12.01	70.00
7.	T7-20 cm X 1.0 cm	63.30	7.20	5.66	50.00
8.	T8-20 cm X 1.5 cm	58.66	5.23	4.63	52.30
9.	T9-20 cm X 2.0 cm	63.33	4.52	4.40	58.20
	Grand Mean	66.31	6.31	6.16	56.70
	SE.d	0.68	0.34	1.34	0.64
	C.D (0.05)	1.43	0.72	2.83	1.36

Table 3: Influence of rooting media on clonal propagation of *Dalbergia sissoo*

S. No	Treatments	Rooting %	Number of primary Roots	Root length per cutting	Survival %
1.	Sand: Soil: VAM(2:1:1)	86.00	36.66	14.28	88.00

Standardization of cutting size

To standardize the cutting size, the semi hardwood stem cuttings were collected from five year old mother tree and based on length, they were grouped into small (10 cm), medium (15 cm) and large (20 cm) sizes.

Treatments for standardizing cutting size are as follows,

T1-10 cm X 1.0 cm
T2-10 cm X 1.5 cm
T3-10 cm X 2.0 cm
T4-15 cm X 1.0 cm
T5-15 cm X 1.5 cm
T6-15 cm X 2.0 cm
T7-20 cm X 1.0 cm
T8-20 cm X 1.5 cm
T9-20 cm X 20 cm

Standardization of rooting media

The IBA solution dipped stem cuttings were planted in different rooting media compositions, cuttings were placed inside low cost ploy tunnels in the hardening chamber. The poly tunnel having warmer temperature of 25-35 °C and humidity maintained around 70-80% with intermittent spray water. Forty-five cuttings in each treatment with three replications were placed. Treatments for standardize rooting media is Sand: Soil: VAM (2:1:1). The data on various rooting parameters was recorded by observing the cutting from each replicate of each treatment after 6 weeks of treatment. The final observations on root initiation, root number per cutting, root length per cutting and survival percentage were recorded after 12 weeks of the experiment. The destructive sampling was done in five randomly selected seedlings for each treatment per replication and then mean was worked out. Data were subjected to analysis of variance (ANOVA) and F-tests for significance as per the procedure devised by Panes and Sukhatme (1967). Critical difference values were calculated for comparing the treatment means at p= 0.05.

Results and Discussions

The data observed in the present study revealed that rooting hormone IBA proved effective on root initiation and growth of adventitious roots of stem cuttings of *Dalbergia sissoo* with in 15 days of treatment and planting. After 12 weeks of the experiment, it has been observed that amongst all the six treatments tried, IBA at 4000 ppm (T4) was the most effective for root initiation. The analysis of variance is presented in table 1, which clearly shows that all the rooting parameters *viz*., rooting percent, root numbers per cutting, root length per cutting and survival percent. It therefore revealed that treatment T4 at 4000 ppm is the best and most efficient hormonal concentration in simulation of higher rooting (86.60%), root number per cutting (56.66), root length per cutting (15.77) and greater survival (86.60%) of stem cuttings of *Dalbergia sissoo*. Moderate concentration of IBA at 4000 ppm could be better than higher (IBA at 5000 ppm) and lower (IBA at 1000 ppm) concentrations. The rooting was very slow and not profuse with T6 (Control). So in order to induce the profuse rooting, treatment of cuttings with IBA at 4000 ppm in particular is found to be beneficial. Exogenous application of IBA may have activated carbohydrate metabolism for release of energy. While protein and peroxidase activity were necessary for cell division and differentiation during adventitious root primordium initiation and development in the rooting zone of cuttings. Palanisamy (1997) [5] described a positive relationship between carbohydrate content in the cuttings with rooting and root numbers. The rooting of stem cuttings varied to a great extent on seasons of cuttings, stem diameter, age of the mother tree, medium of propagation and type and concentration of auxins (Nanda, 1970; Harsh and Mathana, 1985 and Hartmann *et al* 1997) [9, 1, 2]. The exogenous application of hormones had been reported earlier in *Acacia albida* (Nada *et al.* 1969) [8], *Casuarina equisetifolia* and *Gmelina arborea* (Parthiban *et al.*, 1999) [11] and *Ceiba pendanadra* (Rajendran *et al.*, 2002) [12]. The effect of IBA on rooting of stem cuttings was investigated in *pongamia pinnata* Kumaran *et al.*, (2010) [6]. In this context, the present investigation was also carried out for propagation of stem cuttings of *Dalbergia sissoo* using various sizes of cutting. Out of nine cutting sizes tried, 15 cm length with 2.0 cm diameter cuttings reigned supreme in terms of rooting percent (75.81%), root numbers per cuttings (16.1), root length per cuttings (12.01 cm) and also recorded higher value in terms of percent survival (78.80%) of cuttings(Table-2). Similar result was reported by Nautiyal *et al.*, (1994) [10] stating that cuttings taken from lower part of the crown could be rooted to the tune of 60 percent on treatment which corroborated the findings of the present investigation. This is likely to be due to species differences or on an account of the metabolic state of the tissue (Nanda *et al.*, 1969) [9]. The rooting potentiality and its subsequent growth are also affected by the location from which the cuttings are taken (Hartmann *et al.*, 1997) [2] Zarkaria and Ong (1982) [15] obtained a higher rooting percentage from basal stem cuttings in *Gmelina arborea* seedlings which was assigned due to large diameter with more stored carbohydrates for root and shoot. Kumaran *et al.*, 2010 [6] reported that 15 cm length and 2.0 cm diameter size cutting was performed well. The role of rooting media in inducing rooting and developing healthy seedlings in stem cutting is well known. An ideal rooting media should be able to provide enough moisture and nutrients for root initiation and further development besides preventing the desiccation of the cut ends. The result in table-3, indicated that polythene bags containing Sand: Soil: VAM (2:1:1) media markedly improved rooting 86 percent, 36.66 Number of primary Roots, 14.28 cm Root length per cutting and88.00 Survival percent.

Conclusion

It is concluded that in long cuttings of the size of 15 cm X 2.5 cm *Dalbergia sissoo* dipped with IBA 4000 ppm planted in Sand: Soil: VAM (2:1:1) is suitable for enhancing rooting and survival percent of stem cuttings. Such knowledge will aid propagators, geneticists and tree improvement specialists in selecting treatments that reduce propagation costs by maximizing rooting success. Hopefully the present studies will also pave the way to develop clones of superior trees of *Dalbergia sissoo* used for nitrogen fixation, fodder in agroforestry or large scale timber purposes.

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