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SPROUTING PERCENTAGE OF *Dalbergia sissoo* AS AFFECTED BY SIZE OF CUTTING AND DIFFERENT GROWTH HORMONES

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Quantitative study was done on the shoot cuttings of *Dalbergia sissoo* (Shisham) in the Forestry Experimental Area, University of Agriculture Faisalabad. Shoot cuttings of three different sizes 2", 4" and 6" were prepared and were treated with root initiating hormones IBA, IAA and NAA. Hormones were applied in two concentrations 200mg/L and 400 mg/L. Best sprouting percentage of 85.38% was found in 6" cutting in control. Best sprouting percentage with the application of hormones was obtained with IAA 200mg/L which was 85.15%. It was found that hormones of concentration 400 mg/L have a negative effect on sprouting percentage of cutting with lower biomass. Consequently, the sprouting percentage in the cuttings of 2" and 4" size was higher 43.30% and 83.35% in the control. In 2" and 4" cutting treated with 400mg/L, sprouting percentage was 2.167% and 13.10% respectively.

Keywords: *Dalbergia sissoo* cutting, IAA, IBA, NAA, Sprouting Percentage.

INTRODUCTION

In Pakistan, Shisham (*Dalbergia sissoo*) is widely grown as an Agro-forestry tree. Its common names are Shisham, Tahli and Rose wood. It belongs to the family Leguminosae. Sub-order is Papilionodeae. It can grow from medium to a large sized tree up to 30 m. It is deciduous in nature. Branches are spreading type. The trunk is usually crooked. The leaves are compound with 3-5 leaflets on an 8cm stalk. Flowering occurs in groups and is small in size. The flowers are dull white to pinkish in colour and appear in between March to May. The Pods are small, 5 to 8 cm long and papery. The pods ripen from June to February. There are usually 1 to 4 seeds per pod (Sheikh 1987). Shisham (*Dalbergia sissoo*) tree is native to Pakistan, India and Nepal. Its habitat ranges from sea level to more than 1500 m high. It can stand temperatures from below freezing to nearly 50°C. It can withstand average annual rainfall of 2000mm and 3-4 months of drought (NAS, 1979). Though the tree grows best in well-drained soils, it can grow in slightly saline soils as well. It is found to be native to moist deciduous forest generally growing in soil of pH ranging 5.0 to 8.5. Plant sheds its leaves in November and December but is again full leafed in spring and early summer. It flowers abundantly for one month early in summer and is a favored nectar source. Sometimes found gregarious in alluvial forests and along the rivers in the Sub-Himalayan tract. (C.S.I.R., 1948-1976). Young trees coppice vigorously and reproduce vigorously from suckers. Root suckers transplant satisfactorily in dry climates. Planting should be in spring (March in Pakistan). Stump planting is widely employed in irrigated plantations in Pakistan (Nanda, 1970; Hartmann *et al.,* 1997). Shallow and frequent irrigation or constant flooding is harmful and induces superficial root formation. Depending upon the weather and the condition of plants, 10-15 irrigations are adequate in the first season and 4-6 in the second. Under proper irrigation, (*Dalbergia sissoo)* roots tap the subsoil water within 2 years. Irrigation in later years is required only for supplementing subsoil water supplies (Zakaria and Ong, 1982). Shisham (*Dalbergia sissoo)* is known throughout the farmlands of Punjab due to its high quality wood and its non-competitive nature with the farm crops. Pakistan spent Rs. 10.5 billion on the import of wood and wood products during the year 2000-2001. It earned Rs.1.09 billion by the exports of value added wood products during the same year (Anonymous 2002). As Shisham (*Dalbergia sossoo*) is mainly propagated through seed (sexual type of reproduction), thus the plants produced from such method show considerable variation in their growth pattern and disease susceptibility than the parent plant from which the seed has been collected. This method of propagation also induces irregularity in the seed setting season. With the passage of time and prolonged use of traditional method of propagation (through seed) has increased the risk of disease susceptibility in *Dalbergia* spp. Over the last few years, the population of *Dalbergia sissoo* on the farmlands and along the roadside has showed drastic decline due to a disease called "Shisham Dieback". It is a matter of common observation that huge number of Shisham trees has dried up in cultivated tracts and forest plantations that has greatly upset both the farmers and forest personals. One can witness while traveling hundreds of dried *Dalbergia sissoo* trees along the roads. The reason for the mortality of this tree in such a large number is a disease called "Shisham Dieback". The problem has attained an alarming situation and the menace is spreading into fruit trees as well. It has been observed that trees like kikar (*Acacia nilotica*), poplar (*Populus* spp.), willow (*Salix*

tetrasperma), sufaida (*Eucalyptus camaldulensis*) and bakain (*Melia azedarach*) are also dying on farm lands as well as in canal and road-side plantations. The reports are even more heartening about fruit trees like mango (*Mangifera indica*), Citrus and guava. Many experiments and field trials were conducted to cope with this disease but the solutions found were either very expensive or with a little success. After detailed survey it was generally observed that there are some lush green trees which are still standing erect among those disease affected trees. This suggests that, there are resistant strains of Shisham (*Dalbergia sissoo*) which naturally exists and are having considerable immunity towards this Dieback disease. Cuttings are probably the most important method for starting new plants. A cutting is any detached plant part that, under favorable conditions for regeneration, will produce a new plant identical to the parent plant (David 2003). The present work is an effort to determine the best and easiest method of producing plants of Shisham from branch cuttings. This research will also help in determining the reasonable size for the vegetative propagation of Shisham.

MATERIALS AND METHODS

The experiment was conducted in the Forestry experimental area in University of Agriculture Faisalabad. For that a green house of 10 x 12 x 10 feet dimensions was built.

***Selection of the Mother tree*:** Detailed survey around Faisalabad was conducted in order to select the area, which is most seriously affected by Shisham Dieback. Lush green tree, which was standing in the worse hit area for the disease, was selected as the mother plant. Shoot cutting were then taken from that plant. Selection of the tree was done on the following phenotypic qualities.

i) The age of the tree should be between 20-25 years.
ii) The plant which is selected should be disease free, with healthy leaves and branches.
iii) The selected plant should have more or less straight bole.
iv) The selected tree should be standing among some Dieback affected trees.

***Preparation of the cuttings*:** Approximately one year old branches of thumb thickness were selected. Cuttings of three sizes i.e. 2", 4" and 6" were prepared. While preparing the cuttings care was taken that the bark may not get damaged. Flush cutting was done to avoid the pealing away of the bark. A slanting cut was given on the upper end of the cutting to avoid fungal attack. The cuttings were dipped in bleach solution carefully prepared at a ratio of 1:4 in water.

***Treating with root initiating hormones*:** Three root initiating hormones Indole acetic acid (IAA), Indole butyric acid (IBA) and Naphthalene acetic acid (NAA) were used in two concentrations i-e 100mg/lit. and 200mg/lit. Basal 1/3 portion of the cuttings were dipped in the root initiating hormone solutions for 4 hrs.

***Planting of the treated cuttings*:** Cuttings were planted in polythene tubes (6cm x 18cm). These Polythene tubes were filled with silt and well rotten Farm yard manure in a 3:1 ratio, which acted as the rooting medium.

***Treatments in the experiment*:** Following treatments were applied to the cuttings.

Factor A: Cutting size
C1 (2") Cutting size 2"
C2 (4") Cutting size 4"
C3 (6") Cutting size 6"
Factor B: Growth hormones
G1 (IAA) Indole acetic acid
G2 (NAA) Naphthalene acetic acid
G3 (IBA) Indole butyric acid
Factor C: Hormone concentration
T1
(C) Control
T2 (200) 200mg/L
T3 (400) 400mg/L

PARAMETERS RECORDED

***Sprouting Percentage*:** Sprouting percentage is very important to predict the behavior of the tree if it is to be propagated by asexual method of shoot cutting. Good sprouting percentage is a perquisite for a healthy future crop stand.

RESULTS AND DISSCUSSION

The present work is an effort to determine the best and easiest method of producing plants of Shisham from branch cuttings. The root initiating hormones IBA, NAA and IAA will also be used for the vegetative propagation of *Dalbergia sissoo*. In addition, polythene tubes of smaller size will be used to see its effects on root initiation of the cuttings. This research will help in reducing the size of shoot being used in vegetative propagation. It will also reduce the plant genotypic variability through asexual propagation method and producing true to type plants of Shisham and will help to ensure increased germination percentage by determining the best dose of the root initiating hormones. This research will enable us to produce Die-back resistant planting stock of Shisham. Thus, farmers will again be able to grow Shisham on their farmlands without any fear of Die-back disease.

***Sprouting percentage (%age)*:** Sprouting percentage always plays an important role for a good future crop. If there is more sprouting it means the cultural practices you are doing and planting material used is satisfactory. Environmental conditions are also important for good sprouting. High sprouting percentage is the primary requirement for a good nursery business (Ishtiaq *et al*. 1995). Sprouting percentage is very important to predict the behavior of the tree if any species is to be propagated by asexual method of shoot cutting. Good sprouting percentage is a perquisite for a

healthy future crop stand. Cuttings were observed very carefully and after 30 days sprouting percentage was calculated (Mebrahtu *et al*. 1990)

Analysis of variance for the sprouting percentage are given table-1 and the comparison of means for the factors like cutting size (A), Growth hormones (B) and Hormone concentration (C) are given in tables 1, 2 and 3 respectively. The interaction between the factors Cutting size and Hormone concentration (AC), Cutting size and Growth hormone (AB) and Growth hormone and Hormone concentration (BC) are given in the tables 4, 5 and 6 respectively. Analysis of variance (Table-1) for the sprouting percentage shows that all the factors A, B and C along with their interactions AB, AC and BC were found to be significant. Factor A, C and their interaction AC were found to be highly significant.

It is evident from the mean table (Table-1) and the graph (Fig-1) for the factor A, that the sprouting percentage was highest for the C3 (6") which was (72.10 %). It was followed by the C2 (4") and C1 (2") which were (42.89) and (18.77) respectively. Mean table (Table-2) and the graph (Fig-2) for the factor B shows that growth hormone G1 (IAA) was found to be the best showing (51.38 %) sprouting. Lowest sprouting percentage was recorded in G2 (NAA) which was (34.32 %) and (48.06 %) sprouting was recorded in G3 (IBA). Mean table (Table-3) and the graph (Fig-3) for the factor C shows that the shoot cutting which were not treated with any growth hormones T1 (C) showed best sprouting percentage of (74.01 %). It was followed by T2 (200) and T3 (400) with (34.71 %) and (25.03 %) sprouting. Interaction table for the means (Table-4) and the graph (Fig-4) of the factor AB that the highest sprouting percentage was in the C3 (6") with G1 (IAA) which was (75.15 %) and the lowest was recorded with C1 (2") with G3 (IBA) which was (16.43 %). Interaction table for the means (Table-5) and the graph (Fig-5) between the Factors AC that the best sprouting was in the C1 (6") with T1 (C) which was (85.38 %) and the worst was recorded in C1 (2") with T3 (400) which was (2.167 %). Table of interaction of the means (Table-6) and the graph (Fig-6) between factors BC shows that the sprouting percentages in two treatments i.e. with G3 (IBA) and T1 (C) and with G2 (NAA) and T1 (C) was (77.72 %). Least sprouting percentage was recorded with G2 (NAA) and T3 (400) which were (6.433 %). From the results of the sprouting percentage it is clear that 6" cutting under control gave the best result with T1(C) (85.38%) sprouting. Overall 6" cutting was found best in all the treatments but it sprouted maximum in control. 4" cuttings were also good in control but showed less sprouting than 6". 2" cutting showed low sprouting in control and it was even lower with 400mg/L of hormone concentration. Similar results were also found best by the other researchers like (Maria *et al*. 2003), (Wang *et al*. 1997), (Edson *et al*. 1991)

Table 1: Mean table for the factor A:

	C1 (2")	C2 (4")	C3 (6")
Means	18.77	42.89	72.10

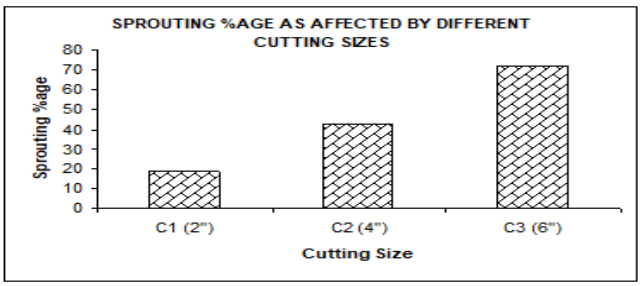

Figure 1: Sprouting percentage as affected by different cutting sizes.

Table 2: Mean table for the Factor B:

	G1 (IAA)	G2 (NAA)	G3 (IBA)
Means	51.38	34.32	48.06

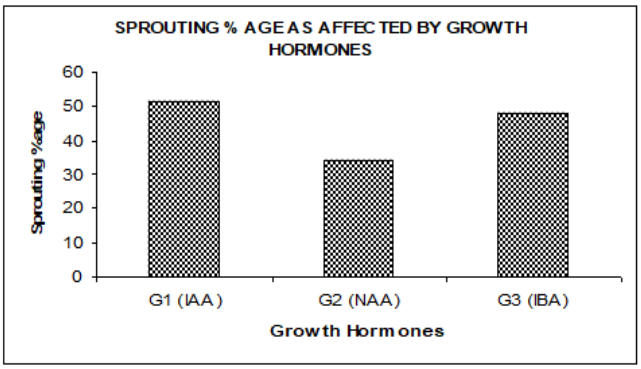

Figure 2: Sprouting percentage as affected by growth hormones.

Table 3: Mean table for the factor C:

	T1 (C)	T2 (200)	T3 (400)
Means	74.01	34.71	25.03

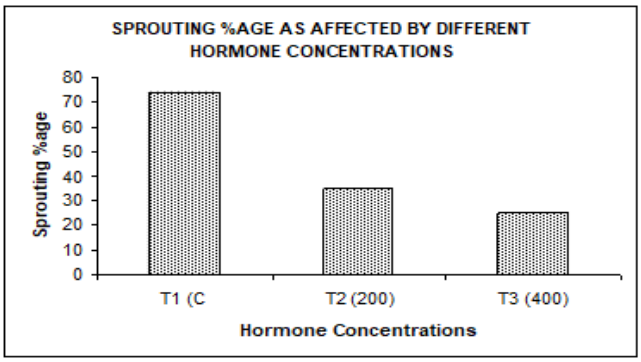

Figure 3: Sprouting percentage as affected by different hormone concentrations.

Table 4: Interaction table between the factors AB:

	C1 (2")	C2 (4")	C3 (6")
G1 (IAA)	23.27	47.66	85.15
G2 (NAA)	16.60	42.23	83.20
G3 (IBA)	16.43	38.77	47.60

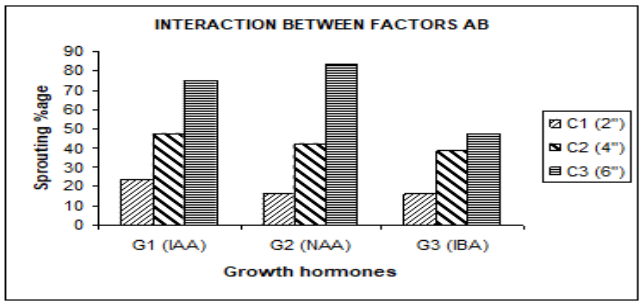

Figure 4: Interaction of factors AB

Table 5:Interaction table for the factors AC:

	C1 (2")	C2 (4")	C3 (6")
T1 (C)	43.30	83.35	85.38
T2 (200)	10.83	22.21	71.10
T3 (400)	2.167	13.10	59.83

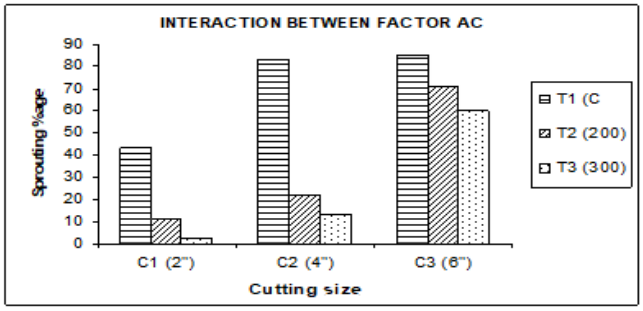

Figure 5: Interaction between factors AC

Table 6: Interaction table for the factors BC:

	IAA	NAA	IBA
Control	66.60	77.72	77.72
200mg/L	48.88	18.82	36.44
400mg/L	38.65	6.433	30.01

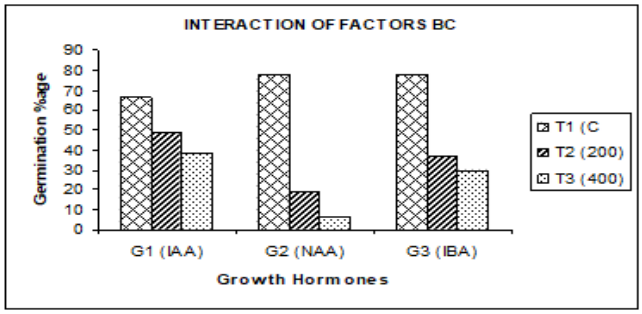

Figure 6: Interaction between factors BC:

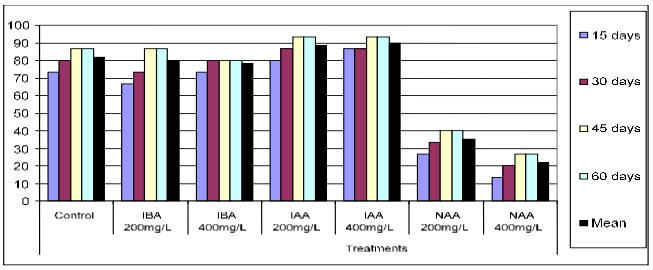

Figure 7: Sprouting Percentage of 6" shoot cutting of *Dalbergia sissoo.*

The table shows that the sprouting percentage in the shoot cutting treated with IAA 400mg/L is highest 89.99%. It is followed by the cutting treated with IAA 200mg/L with 88.33% sprouting. The control, which was not treated with any hormone showed a sprouting percentage of 81.66%. After forty five days the sprouting percentage was same 93.33% in both doses of IAA as compared to that of control with 86.66%. It is clear that among the growth hormones applied, IAA was found to be the most effective. IBA was second and lastly NAA which showed poor sprouting percentage as compared to other with 21.66% sprouting.

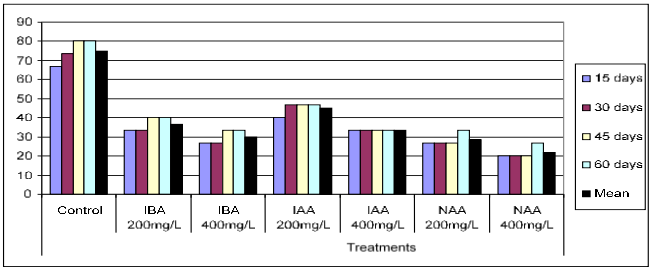

Figure 8: Sprouting Percentage in 4" shoot cutting of *Dalbergia sissoo*

In 4" cutting, best sprouting percentage was observed in control, which was 74.99%. Among the treated cutting, the cuttings which were treated with IAA 200mg/L showed the best results with 44.99% sprouting. Highest mortality was observed in NAA 400mg/L in which 78.44% of cuttings died. The reason for the low sprouting percentage in treated cutting as compared to control was the high concentration of growth hormone as compared to the cutting biomass.

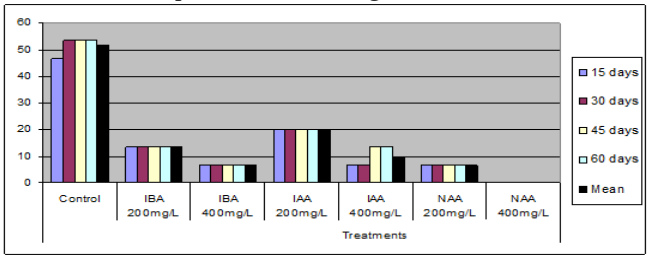

Figure 9:Sprouting Percentage of 2" shoot cutting of *Dalbergia sissoo.*

Table 7: Sprouting Percentage of 6" shoot cutting of *Dalbergia sissoo* .

Time interval	Treatments						
	Control	**IBA 200mg/L**	**IBA 400mg/L**	**IAA 200mg/L**	**IAA 400mg/L**	**NAA 200mg/L**	**NAA 400mg/L**
15 days	73.33	66.66	73.33	80.00	86.66	26.66	13.33
30 days	80.00	73.33	80.00	86.66	86.66	33.33	20.00
45 days	86.66	86.66	80.00	93.33	93.33	40.00	26.66
60 days	86.66	86.66	80.00	93.33	93.33	40.00	26.66
Mean	**81.66**	**79.99**	**78.33**	**88.33**	**89.99**	**34.99**	**21.66**

Table 8: Sprouting Percentage in 4" shoot cutting of *Dalbergia sissoo* .

Time interval	Treatments						
	Control	**IBA 200mg/L**	**IBA 400mg/L**	**IAA 200mg/L**	**IAA 400mg/L**	**NAA 200mg/L**	**NAA 400mg/L**
15 days	66.66	33.33	26.66	40.00	33.33	26.66	20.00
30 days	73.33	33.33	26.66	46.66	33.33	26.66	20.00
45 days	80.00	40.00	33.33	46.66	33.33	26.66	20.00
60 days	80.00	40.00	33.33	46.66	33.33	33.33	26.66
Mean	**74.99**	**36.66**	**29.99**	**44.99**	**33.33**	**28.32**	**21.66**

Table 9: Sprouting Percentage of 2" shoot cutting of *Dalbergia sissoo* .

Time interval	Treatments						
	Control	**IBA 200mg/L**	**IBA 400mg/L**	**IAA 200mg/L**	**IAA 400mg/L**	**NAA 200mg/L**	**NAA 400mg/L**
15 days	46.66	13.33	6.66	20	6.66	6.66	-
30 days	53.33	13.33	6.66	20	6.66	6.66	-
45 days	53.33	13.33	6.66	20	13.33	6.66	-
60 days	53.33	13.33	6.66	20	13.33	6.66	-
Mean	**51.66**	**13.33**	**6.66**	**20**	**9.99**	**6.66**	**-**

The table shows that there was 51.66% sprouting in control. It was followed by IAA 200mg/L with 20% sprouting and IBA 200mg/L with 13.33% respectively. The sprouting percentage in the cuttings treated with NAA 400mg/L was zero. This observation strengthens the idea of negative impact of the high concentrations of growth hormones on the cuttings with low biomass. The hormones concentrations of 200mg/L and 400mg/L proved to be lethal for the shoot cutting of lower biomass like in case of 2" cuttings.

Vegetative propagation techniques have already been used for maintaining genetic superiority and increasing productivity of many economically important tree species (Rafiqul, 2008). The possibility of vegetative propagation of cuttings using mist propagation chambers and rooting hormones from adult *D. sissoo* trees was investigated by Puri and Verma (1996). Husain (2004) studied softwood nodal shoot cuttings from hedge gardens by rooting these in a mist chamber, while the seasonal effect in the rooting of D. sissoo cuttings was observed by Rana *et al*. (1987). Similarly, the effect of light, rooting media and age of mother trees on the rooting ability in *D. sissoo* was studied by Singh *et al*. (2011). The genetic constitution or source of cutting was also significantly affected by the rooting potential in *D. sissoo* (Singh and Bhatt, 2009).

***Conclusion*:** The statistical figures given represent that 6" cutting size is the best for vegetative propagation of *Dalbergia sissoo*. It is also found that IAA with a concentration of 400mg/L is best growth hormone for Dalbergia cuttings. As *Dalbergia sissoo* is hard wood specie unlike *Morus alba*, in which 2" cutting was recommended (W. Irfan 2005). The statistical figures given represent that 4" cutting size is the best for vegetative propagation of *Dalbergia sissoo*. It is also found that IAA with a concentration of 200mg/L is best for the vegetative propagation Dalbergia cuttings. According to the results one can not neglect 4" cutting as its results were found non-significant in three parameters with the results of 6" cuttings. So, from the above results 4" cutting is recommended for it vegetative propagation of *Dalbergia sissoo*. The results have shown that although the sprouting percentage in 2" control was higher than the cutting treated with hormones but still there is a clear difference between the sprouting percentages of 6" control which is 83.38% and 4" which was 83.35% respectively.

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