

**STUDIES ON VEGETATIVE PROPAGATION TECHNIQUES IN  
*Diospyrosebenum* J. KOENIG: AN IMPORTANT  
ENDANGERED TREE SPECIES**

**VILASKUMAR  
ID NO. MF3TAF033**

***DEPARTMENT OF SILVICULTURE AND AGROFORESTRY  
COLLEGE OF FORESTRY, PONNAMPET  
UNIVERSITY OF AGRICULTURAL AND HORTICULTURAL SCIENCES,  
SHIVAMOGGA -577225***

**2019**

**STUDIES ON VEGETATIVE PROPAGATION TECHNIQUES IN  
*Diospyrosebenum* J. KOENIG: AN IMPORTANT  
ENDANGERED TREE SPECIES**

**VILASKUMAR  
ID NO. MF3TAF033**

Thesis submitted to the

**UNIVERSITY OF AGRICULTURAL AND HORTICULTURAL SCIENCES,  
SHIVAMOGGA**

In partial fulfilment of the requirements  
for the award of the degree of

***MASTER OF SCIENCE***

In

**DEPARTMENT OF SILVICULTURE AND AGROFORESTRY**

**Shivamogga**

**November, 2019**

DEPARTMENT OF SILVICULTURE AND AGROFORESTRY  
COLLEGE OF FORESTRY, PONNAMPET  
UNIVERSITY OF AGRICULTURAL AND HORTICULTURAL SCIENCES  
SHIVAMOGGA

CERTIFICATE

This is to certify that the thesis entitled "STUDIES ON VEGETATIVE PROPAGATION TECHNIQUES IN *Diospyros ebenum* J. KOENIG: AN IMPORTANT ENDANGERED TREE SPECIES" submitted in partial fulfillment of the requirements for the award of the degree of **MASTER OF SCIENCE (FORESTRY) in SILVICULTURE AND AGROFORESTRY** to the College of Forestry, Ponnampet, University of Agricultural and Horticultural Sciences, Shivamogga is a bonafide record of research work carried out by **VILASKUMAR, ID NO. MF3TAF033** (vilasc05@gmail.com) during the period of study in this university under my guidance and supervision and no part of this thesis has previously formed the basis for the award of any other degree, diploma, associateship, fellowship or any other similar titles.

Shivamogga  
November, 2019



(MAJOR ADVISOR)

APPROVED BY:  
Chairman:

(MAHESWARAPPA V)

Members:

1.

(RAMAKRISHNA HEGDE)

2.

(R. N. KENCHARADDI)

3.

(VASUDEV LAMANI)

## **ACKNOWLEDGEMENT**

*It always is a nostalgic feeling whenever one glance back to the days of hard work, tensions and the need of the hour of excel. One would not achieve whatever he is now, without all the help, encouragement and the wishes of the near and the dear ones. Teachers, parents, friends and well-wishers are an integral part of this. I owe them a lot and it always is a difficult task expressing and putting into words the sense of gratitude I feel towards them. I wish to remember and acknowledge all who made this thesis possible.*

*With deep respect I express my heartfelt gratitude and unforgettable indebtedness to my esteemed teacher and Chairperson **Dr. Maheswarappa V**, Assistant Professor, Dept. of Silviculture and Agroforestry, College of Forestry, Ponnampet, whose vision was the guiding light of this investigation. His timely suggestions, immaculate guidance, resilient mental support, persistent cooperation, critical evaluation and valued comments and everlasting patience during the study period made my research work an easy task. I express my heartfelt and sincere thanks to him. It was indeed a great privilege of mine to be one of his students.*

*I would like to acknowledge the academic and technical support provided by the **University of Agricultural and Horticultural sciences, Shivamogga** and my esteemed institution, College of Forestry, Ponnampet, in the successful completion of my research work.*

*I place on record my sincere and profound gratitude and reverence to prof., **Ramakrishna Hegde** **Dr. R. N. Kencharaddi**, Asst. Prof., **Mr. VasudevaLamani**, Asst. Prof., for their immense contribution and support during the course of investigation. I owe my sincere thanks to **Dr. C. J. Kushalappa**, Dean (Forestry) who provided the required infrastructure for me during the execution of the work and the teaching faculty and staff of College of Forestry, Ponnampet for their valuable help to carry out my research. My sincere and special thanks to Prof, **G.M. Devagiri**, **Mr. Veerendra M.** (RFO)*

*Dr. AnilkumarKhaple Asst. Prof. on contract basis, Mr.Nanayya K, Asst. Prof. on contract basis, and Ms.SupriyaSalimath Asst. Prof. on contract basis, who helped me during the study.*

*I cannot forget the boundless backing given by my dear friends and batchmatesJnanashekahar, Anil, Naresh, Chandana, Diana, Swarna, Roopa, Poojashri, I am awfully grateful to my seniors Naman, Vinayak, Sunil Bhavya, Nagamani, Nandini, Manoj , and Manuduring my study. for their kind guidance throughout this period and with immense pleasure I thank them for being who they are.*

*I would also like to thank my juniors Bharath, Krishnappa, Akshay, Sachin, Kiran, Naveen, HaleshMadhuprasad, Suraj, Basavaraj, Ashwath, Akash, Kshama for their endless support during the study.*

*At this juncture, my heartfelt respect for my family where I come from and the source of my pride. Words fall short to express my special thanks to my Father Mr. Ashok Patil, MotherMs. Anita Patil, Grand fatherChannappa M and Grand motherKalavati C. aunty kavyamamadgi, Uncle Kuberraj, brothers and sisters SuryaprakashPatil, Shivaprasad, Sai Prasad, Sachin, Harsha and Aditya, Swapna, ArathiPatilSatyashree, and Meghapatil, Dimpri, Nagasinchana and Sathvik for their boundless love, selfless prayers, abundant affection and continuous encouragement throughout my studies.*

*Ending is inevitable for any kind of work through acknowledging as endless. I end by saying endless to all those whom I am unable to recall here and also those whom I might have left unknowingly.*

Ponnampet



(Vilaskumar)

**Studies on Vegetative propagation Techniques in *Diospyros ebenum* J. Koieng: An Important Endangered Tree Species**

**ABSTRACT**

*Diospyros* is a large genus belongs to family Ebenaceae among which *Diospyros ebenum* is a valuable tree species native to India and Sri Lanka. The tree species of the family were slow growing in nature which grows up to 30m height. Wood of this species is known as black ebony and resistant to insect attack and fungi and export to China and Europe for furniture and fancy wood. The usage of the species made considerable loss in number and its frequency and categorized endangered as per IUCN. The vegetative propagation techniques plays important role in conserving such species when there is a problem of natural regeneration and recalcitrant seed to store for a year with improper germination envisage an alternate method of propagation. The present study was undertaken with an objective to identify suitable vegetative propagation techniques and determining the different concentration of IBA with air layering and different types of cuttings. The experiment was conducted at College of Forestry, Ponnampet with standard procedures using different concentrations of IBA, vegetative propagation methods viz., air layering, softwood cuttings, semi hardwood cuttings and hardwood cuttings. The findings of the research indicated the higher mean root number (6.16) in air layering treated with 2000 ppm of IBA. Rooting success was 9.48 % in semi hardwood cuttings treated with 2000ppm of IBA with fresh shoot and root weight of 0.46g and 0.43g respectively.

Department of Silviculture and Agroforestry  
College of Forestry, Ponnampet  
Ponnampet- 571216  
November - 2019



Vilaskumar

[vilascof05@gmail.com](mailto:vilascof05@gmail.com)



Dr. Maheswarappa. V

[cv\\_mahesh@rediffmail.com](mailto:cv_mahesh@rediffmail.com)

ಅಳವಿನಂಚಿನಲ್ಲಿರುವ ಡಯೋಸ್ಟೈರೋಸ್ ಎಬೆನಮ್ ಜಿ. ಕೋಯೆಂಗ್ ಸಸ್ಯಾಭಿವೃದ್ಧಿ ಬಗ್ಗೆ ಅಧ್ಯಯನ

## ಸಾರಾಂಶ

ಡಯೋಸ್ಟೈರೋಸ್ ಎಬೆನಶಿಯ ಕುಟುಂಬಕ್ಕೆ ಸೇರಿದ ಒಂದು ಪ್ರಭೇದವಾಗಿದ್ದು ಇದರಲ್ಲಿ ಡಯೋಸ್ಟೈರೋಸ್ ಎಬೆನಮ್ (ಕರಿಮರ) ಭಾರತ ಮತ್ತು ಶ್ರೀಲಂಕಾ ದೇಶಗಳಿಗೆ ಸ್ಥಳೀಯವಾಗಿದೆ. ಈ ಕುಟುಂಬದ ಮರಗಳು ನೈಸರ್ಗಿಕವಾಗಿ ೩೦ಮೀ ಎತ್ತರ ಬೆಳೆಯುತ್ತವೆ. ಅತ್ಯಂತ ಅಮೂಲ್ಯವಾದ ಈ ಕರಿ ಮರವನ್ನು ಕಪ್ಪು ಎಬೊನಿ ಎಂದು ಕರೆಯಲಾಗುತ್ತದೆ. ಕೀಟಗಳ ದಾಳಿ ಹಾಗೂ ಶಿಲೀಂಧ್ರಗಳಿಗೆ ನಿರೋಧಕವಾಗಿರುವ ಈ ಮರವನ್ನು ಪೀಠೋಪಕರಣಗಳಿಗಾಗಿ ಹಾಗೂ ಅಲಂಕಾರಿಕ ಮರವಾಗಿ ಚೀನಾ ಹಾಗೂ ಯುರೋಪ್ ದೇಶಗಳಿಗೆ ರಫ್ತು ಮಾಡಲಾಗುತ್ತದೆ. ಇದರಿಂದಾಗಿ ಈ ಮರಗಳ ಸಂಖ್ಯೆ ಕಡಿಮೆಯಾಗಿರುವುದರಿಂದ ಐಯುಸಿನ್ ಈ ಪ್ರಭೇದವನ್ನು ಅಳವಿನಂಚಿನಲ್ಲಿರುವ ಮರವೆಂದು ವರ್ಗೀಕರಿಸಲಾಗಿದೆ. ಈ ಮರಗಳ ಅನುಚಿತ ಬೀಜಾಂಕುರದಿಂದ ಗಿಡವಾಗುವುದು ಕಡಿಮೆ. ಇದು ಸಸ್ಯಾಭಿವೃದ್ಧಿಗೆ ಹಿನ್ನಡೆಯಾಗಿದೆ. ಈ ನಿಟ್ಟಿನಲ್ಲಿ ಮರದ ಕಾಂಡದ ಕಡ್ಡಿಗಳ ಹಾಗೂ ಗೂಟ ಕಟ್ಟುವಿಕೆಯ ಮೂಲಕ ಸಸ್ಯಾಭಿವೃದ್ಧಿ ಮಾಡುವ ಪ್ರಯತ್ನವನ್ನು ಈ ಅಧ್ಯಯನದಲ್ಲಿ ಮಾಡಲಾಯಿತು. ಇದರಲ್ಲಿ ವಿವಿಧ ರೀತಿಯ ಕಾಂಡಗಳನ್ನು ಹಾಗೂ ಗೂಟಗಳನ್ನು ಹಲವಾರು ಪ್ರಮಾಣದ ಬೆಳವಣಿಗೆ ಪ್ರಚೋದಕಗಳಿಂದ (ಐಬಿಎ) ಉಪಚರಿಸಿ ಅವುಗಳ ಫಲಿತಾಂಶವನ್ನು ಗಮನಿಸಲಾಯಿತು. ಅಧ್ಯಯನದ ಫಲಿತಾಂಶಗಳ ಪ್ರಕಾರ ೨೦೦೦ಪಿಪಿಮ್ ನಲ್ಲಿ ಉಪಚರಿಸಿದ ಗೂಟಗಳಲ್ಲಿ ಅಂದಾಜು ಬೇರುಗಳ ಸಂಖ್ಯೆ (೬.೧೬) ಹೆಚ್ಚಾಗಿರುವುದು ಕಂಡುಬಂದಿರುತ್ತದೆ. ಇದಲ್ಲದೇ ಅರೆಮೆತ್ತನೆಯ ಕಾಂಡದ ಕಡ್ಡಿಗಳಲ್ಲಿ ಬೇರು ಬಿಡುವ ಯಶಸ್ಸು (೯.೪೮ %) ಹಾಗೂ ತಾಜಾ ಕಾಂಡ (೦.೪೬ ಗ್ರಾಂ) ಮತ್ತು ಬೇರುಗಳ (೦.೪೩ ಗ್ರಾಂ) ಜೀವರಾಶಿಯು ಅತ್ಯಧಿಕವಾಗಿರುವುದು ಕಂಡುಬಂದಿರುತ್ತದೆ.

ಅರಣ್ಯ ಕೃಷಿ ಮತ್ತು ಕೃಷಿ ಅರಣ್ಯಶಾಸ್ತ್ರ ವಿಭಾಗ  
ಅರಣ್ಯ ಮಹಾವಿದ್ಯಾಲಯ, ಮೊನ್ನಂಪೇಟೆ  
ಕೃಷಿ ಮತ್ತು ತೋಟಗಾರಿಕೆ ವಿಶ್ವ ವಿದ್ಯಾಲಯ, ಶಿವಮೊಗ್ಗ  
ನವೆಂಬರ್, ೨೦೧೯

ವಿಲಾಸಕುಮಾರ

[vilascof05@gmail.com](mailto:vilascof05@gmail.com)

ಡಾ. ಮಹೇಶ್ವರಪ್ಪ. ವಿ

[cv\\_mahesh@rediffmail.com](mailto:cv_mahesh@rediffmail.com)

## CONTENTS

Chapter	TITLE		Page No.
<b>I</b>	<b>INTRODUCTION</b>		1-3
<b>II</b>	<b>REVIEW OF LITERATURE</b>		4-16
	2.1	Importance of vegetative propagation in forestry	4
	2.2	Importance of growth regulators	8
<b>III</b>	<b>MATERIAL AND METHODS</b>		17-20
	3.1	Description of the study area	17
	3.2	Experiment on vegetative propagation	17
	3.3	Data collection	18
	3.4	Statistical analysis	20
<b>IV</b>	<b>EXPERIMENTAL RESULTS</b>		21-35
	4.1	Initiation of new shoots by different vegetative propagation techniques and growth regulators	21
	4.2	Initiation of shoots and roots in types of cuttings	21
	4.3	Interaction effect of vegetative propagation method and IBA concentration on production of shoot and roots	24
	4.4	Effect of types of cuttings, concentration of IBA and its interaction on growth regulators	27
	4.5	Biomass of rooted cuttings under different concentration of IBA	29
<b>V</b>	<b>DISCUSSION</b>		36-41
	5.1	New shoots and roots with different vegetative propagation method and growth regulators	36
	5.2	Shoots and roots in softwood cuttings	37
	5.3	Shoots and root in semi hardwood cuttings	37
	5.4	Shoot and root in hardwood cuttings	37
	5.5	Effect of IBA and vegetative propagation methods on shoot and root production	38

	5.6	Effect of IBA and vegetative propagation method on root initiation	39
	5.7	Effect of IBA and softwood, semi hardwood and hardwood cuttings on production of shoots	39
	5.8	Effect of IBA and softwood, semi hardwood and hardwood cuttings on production of rooting	40
	5.9	Biomass of rooted cuttings under different concentrations of IBA	40
<b>VI</b>	<b>SUMMARY</b>		42-44
<b>VII</b>	<b>REFERENCES</b>		45-53

## LIST OF TABLES

<b>TABLE NO.</b>	<b>TITLE</b>	<b>PAGE NO.</b>
4.1	Root number, root length and leaf number in different vegetative propagation techniques and concentration of Indole Butyric Acid	22
4.2	Response of cuttings to different concentrations of Indole Butyric Acid (IBA)	23
4.3	Effect of vegetative propagation methods, different IBA concentration and their interaction on growth attributes	25
4.4	Interaction effect of types of cuttings and IBA on production of shoots and roots	28
4.5	Biomass of soft wood cuttings treated with different concentration of IBA	30
4.6	Biomass of Semi hardwood cuttings treated with different concentration of IBA	32
4.7	Total fresh and dry weight (g) in hard wood cuttings with different concentration of IBA	35

### LIST OF FIGURE

<b>FIG NO.</b>	<b>TITLE</b>	<b>BETWEEN PAGES</b>
3.1	Meteorological observations during the study period from January 2016 to March 2018 at CoF, Ponnampet	18-19

## LIST OF PLATES

<b>FIG NO.</b>	<b>TITLE</b>	<b>BETWEEN PAGES</b>
3.1	Healthy selected <i>Diospyrosebenum</i> tree for the air layering	20-21
3.2	Different types of cuttings used for initiation of sprouts and roots in the study	20-21
5.1	Root and shoot initiation in different concentrations of growth regulator (IBA) in air layering	37-38
5.2	Root and sprout initiation in different concentrations of growth regulator (IBA) in cuttings	39-40

---

---

# **INTRODUCTION**

---

---

## I INTRODUCTION

Tropical forests contain much terrestrial biodiversity, provide food, shelter, health care, protect water and soil resources, store carbon in biomass and maintain the delicate composition of the atmosphere (Neuwinger, 2000). One very important material provided by forests is wood which is used in construction, as fuel, in making furniture and other implements. Tropical timbers are preferred as a source of wood because of their natural durability and good working properties (Miller and Wiedenhoeft, 2002). Despite of these important uses, tropical forests are threatened by unsustainable use of land and resource use (De Capua, 2005). The greatest constraint to forest regeneration projects is the lack of good planting material.

Global forest cover is in a continuing state of change. According to the recent report on global forest resource assessment, some countries are showing reduction in total area under forests whereas some other countries are showing positive trend (FAO, 2015). Maintaining intact forests, expanding forest area and restoring forests have been promoted because of their perceived benefits. Even though, the ecological services provided by the forest are getting attention but the preliminary focus has always been given to the production aspect. Many countries, including India, are having some restriction by law to remove wood from the natural forest. Hence, forest plantations have become the major source for meeting the increased demand for wood and other forest products. The success of a plantation depends majorly on the seed source as well as the method of propagation. The only way to harness the desirable variation in the nature is through vegetative propagation.

The common way by which plants regenerate naturally is propagation by seed. For research and rapid improvement of undomesticated species, however, artificial regeneration by vegetative propagation methods offer several advantages. Individuals may be recognized within a population that produces a higher quality of the desired products or services. It would, therefore, be advantageous to propagate these individuals vegetatively to ‘capture’ the genetic variation expressed which may otherwise get lost or diluted during sexual propagation.

Vegetative propagation methods have been developed and used for centuries. Especially in temperate regions, vegetative propagation techniques have been standardized for fruit yielding tree species. Further, particular methods of vegetative propagation have been developed for different species. Tropical fruit species have been subjected to vegetative propagation in a number of cases that have found a lucrative export market. Tropical timber species have also been cloned, mainly for plantations where uniform trees are needed. Many indigenous trees with potential high monetary benefits are only used from natural stands.

*Diospyros* is a large genus belongs to family Ebenaceae which is native to India and Srilanka. It is slow growing medium sized tree grows up to 30m tall and up to 90cm in diameter with bole straight, and with buttresses up to 2m height from ground dense crown with scaly bark surface, fissured, black to grey-black in colour (Finkel *et al.*, 2002). In traditional medicine and Holbrook *Diospyros ebenum* fruits are used in medicine for a Snake bite, diarrhoea, biliousness, ulcer and eaten in times of famine (Topo, 1997). The wood is difficult to seasoning and work by hand. Wood of this species is known as black ebony. It is resistant to Insect attack and fungi and very durable. It is mainly exported to China for furniture and to Europe as fancy wood. It finds in sports goods, musical, mathematical instruments, ornamental carvings and turnery.

Considering the importance of the species there has been large scale harvesting of the species since ancient times and the species has been categorized as endangered as per IUCN (IUCN, 2010).

A comprehensive review of benefits, risks, risk management and technical problems of vegetative propagation has been studied by Burdon and Aimers-Halliday (2003). The main benefits include capture of non-additive genetic gain, uniformity of the product and tailoring of the product to the growing site. The risks include genetic uniformity, biotic risks, inability in adapt to climate change and changing markets. Vegetative propagation can be achieved by various means viz; grafting, air layering rooting of cuttings, coppicing or *in vitro* propagation. Plant hormones play an important role in the development of callus and the differentiation into new roots or vascular tissues. They are chemical substances, which occur naturally in plants in very low concentrations. In addition to the naturally occurring (endogenous) hormones, there are several synthetic or natural substances that have similar effects.

Physiological aspects such as type of cuttings, aspects of etiolation of shoots in air layering media and most importantly growth regulators have all known to influence adventitious rooting either from cuttings or from air layered branches. Discovery of growth regulating substances such as auxins and cytokinins revolutionized the industry of propagation during mid-thirties.

The influence of these aspects has to be studied in relation to a particular species in question to evaluate their efficacies in *Diospyros ebenum*. The regeneration in natural forests is very scanty and influenced with pre and post fungal attack, wild animal damage, difficult in storing the fruits for longer period and sometimes difficult natural bed for its proper germination envisages for vegetative propagation studies. Hence, the present study on standardization of vegetative propagation methods and hormonal treatment of *Diospyros ebenum* through cuttings and air layering was undertaken with the following objectives.

1. To identify suitable vegetative propagation techniques for *Diospyros ebenum* tree species.
2. To determine the appropriate combination of growth regulators for different vegetative propagation techniques.

---

---

# **REVIEW OF LITERATURE**

---

---

## II REVIEW OF LITERATURE

Present study titled “**Vegetative propagation techniques in *Diospyros ebenum* J. Koenig-An endangered tree species**” was carried out with an overall objective of identifying the suitable vegetative propagation techniques and determining the appropriate combination of growth regulators for each of the vegetative propagation technique in *Diospyros ebenum*. This chapter presents the literature reviewed relevant to the present investigation.

### **2.1 Importance of vegetative propagation in forestry**

Propagation is one of the important aspects in forestry. Vegetative propagation methods such as stem cuttings, air layering, budding and grafting are being widely followed to multiply plants of desired characteristics and to maintain their purity for commercial exploitation in many forestry species.

The art of vegetative propagation method has gained a major importance in the field of forestry. Many of the forestry species which are found to be difficult propagate by seed are made to root easily by use of root inducing hormones and providing suitable environmental conditions. The relevant studies on vegetative propagation by stem cuttings and air layering in different forestry species in India and elsewhere are reviewed here so as to get a thorough knowledge about existing technology on vegetative propagation.

Dawson and Were (1997) opined that vegetative propagation methods have been developed and used for centuries. The most important vegetative propagation techniques for tree species are stem or root cuttings, layering and micro propagation techniques.

#### 2.1.1 Identification of suitable vegetative propagation techniques in *Diospyros ebenum*

*Diospyros ebenum* is a slow-growing medium-sized tree grows up to 20-25 meters tall, which belongs to the family *Ebenaceae* commonly known as “Kari mara” which is native to India and Srilanka. It is known for its black coloured wood since ancient times. Timber of *Diospyros ebenum* is said to produce the best commercial black ebony. It is resistant to insect attack and fungi and highly durable. Due to high exploitation of this species for its timber it is listed as a rare and endangered tree species. Lack of quantitative data on the baseline information on the population status of this tree species in natural forest is a barrier plan for the future conservation and management hence the present investigation carried on density, population structure and regeneration of *Diospyros ebenum* were carried out in the natural forest of Kodagu. The result revealed that the density and basal area was

found to be higher in Makutta. The overall regeneration showed a reverse J shape curve but in regeneration of *Diospyros ebenum* showed lack of regenerates in Class I and in higher classes which may be due to various biotic and abiotic pressures in the natural forest.

In natural condition this species generally regenerate by seeds. But the fallen seeds were subjected to severe insect and wild animal disturbance on regenerating sprouts (Schemerbeck and Naudiyal, 2018). Hence it is prudent to explore the alternate methods for species propagation other than seed.

The value of vegetative propagation in forestry is well. Hence, understood, the transfer of non-additive characteristics through seeds is difficult. However, it is routinely possible through vegetative propagation. Therefore, this can be used for achieving grains, such as growth, which has low heritability. The constrains in this approach pertain to biological problems related to maturation state of donar plant, cutting environments, the condition of cuttings and survival / growth of cuttings.

#### 2.1.2 Vegetative propagation techniques in other tree species

Hassan (1977) and Banik (1984) have stressed the importance of selecting the branch cuttings that have spontaneous *in-situ* rooting and rhizome tips at base. The pre-rooted and pre-rhizomed branch cuttings of *Bambusa balcooa* can be obtained by chopping the clum tops and removing the newly emerging clums and rhizome bud (Banik, 1984). Removal of emerging culms was found to produce better result in pre-rooted and pre-rhizomed branches per bamboo clump as compared to chopping the culm top. These cuttings produce active roots and rhizomes and the survival rate in the field is high (90-100%) and this method is ideal for *D. asper* (Hassan, 1977).

Singh and Srivastava (1982) reported 84 per cent success with inarching and soft wood grafting in the month of July. When the scions of Dashehari were grafted on the seedling restock of Bappakai variety. Patel and Amir (1981) observed that softwood grafting between the third week of May and third week August resulted in 90 to 100 per cent graft take, and it was to 90 per cent between February and May.

Seethalakshmi *et al.* (1983) observed variable rooting frequency from branch cuttings in *Bambusa balcooa*. They observed less rooting frequency (40%) from branch cutting as compared to culm cutting (50%) of 2-3 years old clump.

Gonzales *et al.* (1991) from the Philippines reported that split- culm cuttings can reduce the weight of the planting stock in comparison to those produced whole culm cuttings. They obtained 100 per cent survival with cuttings of *Bambusa blumeana* var. *luzonensis*, *Bambusa phillippinensis* *Bambusa vulgaris* *Bambusa vulgaris* var. *Striata* and *Gigantochola asper*.

Das *et al.* (1997) studied the vegetative propagation in *Heritiera fomes* and *H. littoralis* through rooting in pre - girdled stem cuttings. Girdling of stem cuttings is one of the methods of vegetative propagation to enrich of secure the quality of planting material.

Ahlawat and Singh (2000) have reported rooting from bi-nodal culm cuttings of *B. balcooa*, *Bambusa nutans* and *Bambusa tulda*. The rooting and sprouting varied among the species and treatments used. *Bambusa balcooa* exhibited maximum response (140-160) followed by *Bambusa nutans* and *Bambusa tulda*.

Agnihotri and Ansari (2000) observed that, out of the double node and triple node culm branch cuttings of *B. vulgaris* and *D. strictus*, triple node cuttings were found better for high rate of rooting for large scale production of clonal plants.

Kumar (2000) found 76 per cent rooting in *Acacia mangium* with 2000 ppm IBA. Chandramouli (2001) reported the effect of different concentration of growth promoters on stem cuttings of *Sesbania sesban* and *Sesbania grandiflora* of IBA, NAA, whereas *S. grandiflora* did not root at higher concentration of IBA and NAA. Boric acid was most effective in the induction of rooting and sprouting and most favourable for root and shoot growth in both species. Untreated cuttings showed good response in case of rooting as well as sprouting cuttings.

Thatoi *et al.* (2001) studied the effect of auxins on stem cuttings of mangrove species and 100 per cent sprouting and rooting were achieved in *Pongamia pinnata* with IBA 500ppm while application of 2000 ppm IBA was effective in all over mangrove species. Karoshi and Hegde (2000) reported the highest rooting (80 %) in stem cuttings of *Pongamia pinnata* with IBA 2500 ppm, while it was longest (20 %) in untreated cuttings.

Mukhia (2005) reported that, main branch and sub branch cuttings of sympodial bamboo can be used for propagation and they have adventitious buds that sprout and grow roots with high survival rate.

Singh *et al.* (2002) observed that, culm cuttings exhibited better for adventitious root formation and growth as compared to culm branch cuttings in *B. nutans*. The maximum rooting percentage was observed in medium size cuttings of *Commiphora wightii* of stem branch cuttings which is treated with 500 ppm Indole 3 butyric acid.

Babitha *et al.* (2002) studied the effect of auxin on rooting of stem cuttings of *Givotia rottleriformis* and reported that the cuttings treated with IBA 3000 ppm showed highest rooting (77.8 %) while, IAA 5000 ppm showed lowers rooting (33.3 %) but no rooting was observed in control treatment.

Mishra *et.al.* (2002) studied the effect of IBA on *Acacia nilotica* and neem (*Azadirachta indica*) during the months of February, May and August and observed that callus was formed at the girdled portions of all the air layers, with/without IBA treatments. Adventitious rooting was obtained in the air layers of February and May while, the layers made during August failed to form roots. Even the slight delay in the air layering trails during monsoon period may suppress air layering of branches (1-1.5 cm diameter) of *Acacia nilotica* spp. *A. indica* in mid-August and September and findings showed the rooting percentage ranged from 80 to 85 in 1000 ppm and in 1000 to 1600 treatments during August and September respectively, whereas survival rate of rooted air layers varied from 67 to 100% after two months of planting and rooting was completed in 60± days.

Bhojvaid and Negi (2003) studied the effect of season and IBA on air layering in rudrakshha (*Elecarpus ganitrus*) in pre monsoon, and post monsoon. and Maximum rooting was found with IBA 500 ppm in pre monsoon season, followed by monsoon and post monsoon season, whereas a highly significant variation ( $p < 0.05$ ) was observed within the effect of season and callus formation as well as root initiation.

Basak (2003) tested the possibility of artificial regeneration through vegetative propagation of *Aegiceras corniculatum*. Rooting in air layer was induced with IAA, IBA and NAA however, maximum rooting ability was recorded in the air-layers treated with IBA 2500+NAA 5000ppm with a promising rate of survivability. Untreated air-layers, however did not produce any root.

Bhutani (2003) treated air layering of *Pernettya mucronata* with auxins in July or August and highest rooting percentage and 100 per cent survival of plants in July were found with IBA 1000 ppm while number of primary roots and new layers per cuttings was best with 0.5.

Chauhan *et al.* (2003) propagated *Azadirachta indica* (Neem) and *Melia azadiracht* (Dek) by air layering using different IBA and IAA concentrations and maximum rooting (80%) was recorded with IBA 600 ppm in neem, while IBA and IAA @ 800 ppm recorded same response in dek. IAA 600 ppm recorded the highest number of roots (8) and root length (6.9cm) in neem, while IBA 800 ppm recorded the highest number of roots (8) and root length (7.6cm) in dek.

Krishnan *et al.* (2003) studied the effect of different concentrations of IBA on the rooting of air layers in *Simarouba glauca* and observed that IBA @ 3000 ppm promoted 100% success percent than the control. Application of IBA @ 4000 ppm induced more rooting and survival per cent.

Kochhar *et al.* (2005) studied vegetative propagation of *Jatropha curcas* and *J. grandiflora* through stem cuttings and the accompanying biochemical changes. During the rooting process, many changes took place both at the physiological and biochemical levels and activities of many enzymes were up and down regulated. The initial levels of endogenous auxins and their oxidation (IAA oxidase and peroxidases) played a significant part in the process, being higher in *Jatropha grandiflora* than in *Jatropha curcas*. IAA oxidase activity was involved in both root initiation and elongation. Position of the cuttings on the mother branches also played a significant role in rooting and sprouting. Cuttings made from the middle portion of the mother branches exhibited better rooting compared to the most apical or most basal cuttings. The results supported by the peroxidase isoenzyme analysis the cuttings.

Ruiz and Gracia *et al.* (2005) evaluated the effects of IBA levels on rooting capacity of three type juvenile cuttings (apical, intermediate and basal) of *Gmelina arborea* and reported that the rooting and sprouting (54.9%) capacity than the basal ones (43.7 and 38.3% respectively) with 30% more root formed. IBA applications inhibited rooting capacity of apical cuttings, but stimulated it on interactions and basal cuttings. The highest rooting percentage was obtained apical cuttings without IBA (80%) but no significant differences in carbohydrates content were found among cutting types.

## **2.2 Importance of growth regulators**

Auxins often hasten root initiation, increases the number and percentage of cuttings rooted as well as quality of root produced per cutting. Larsen *et al.* (1997) reported that the most reliable rooting hormone is Indole Buyteric Acid (IBA) as compared to Napthalene Acetic Acid (NAA).

Chimel (1985) reported that two most important Auxins, *viz.*, IBA and NAA have been used widely either singly or in combination for induction of rooting in stem cuttings of various crop species and treatment with auxins has been shown to increase the per cent of rooting, hasten initiation and increase the number and quality of roots in a large number of plant species.

Leakey (2004) opined that growth regulators, especially auxins play a major role in induction of rooting. Typically cuttings treated with auxins root more rapidly and produce more roots, usually with a higher percentage of cuttings rooted. According to him treatments with auxins have been shown to hasten root initiation and increase the number, quality of roots and also percentage of rooting. Indole-3-butyric acid (IBA) is found to be the most effective root promoting auxin. The two most important auxins are IBA and Napthalene Acetic Acid (NAA) which are widely used for rooting in a variety of species.

Auxins help in the accumulation of metabolites, synthesis of new proteins, cell enlargement, cell division and increase in nitrogen content leading to callus formation which in turn grows into roots in plum (Strydem and Hartmann, 1960).

The enhancement of rooting may not be due to actual auxin applied, but perhaps due to transformation of that auxin applied and absorbed by the cuttings. Further, synthetic auxins when applied to the cuttings usually increase the development of existing root primordium (Haissig and Davies, 1984).

Suryanarayana and Venkateshwara (1982) obtained the highest percentage of successful layers and highest number of roots per layer in mango treated with IBA@ 2000 ppm in August, September and October.

Ghosh and Basu (1974) noticed a slight fall in the concentrations of total available carbohydrates till root emergence stage, but subsequent fall in total carbohydrates was more with the onset of root emergence. This fall was greater in IBA and NAA treated cuttings than in control.

### 2.2.1 Effect of growth regulators and stem cuttings on sprout and rooting in Forest species

Gehlot *et al.* (2014) studied the effect of auxin on adventitious root induction in mini-cuttings of *Azadirachta indica*. Different concentrations of IAA, NAA and IBA were applied to the mini cuttings. Among the auxins used (IBA, IAA and NAA), IBA showed better results. 250 mg L<sup>-1</sup> IBA treatment gave significantly best results in terms of rooting (80%) followed by 500 mg L<sup>-1</sup> IBA treatment (60 %).

Bhagat *et al.* (1998) reported that IBA at 4500 ppm exhibited best performance with respect to rooting success (90.83 %) and survival of hardwood cuttings of guava.

In *Malus sylvestris* when IBA was applied to stem cuttings or micro cuttings to stimulate rooting, a part of IBA was converted to IAA and IBA itself enhanced rooting by increasing the internal free IBA, or synergistically by modifying the action of IAA. IBA can enhance tissue sensitivity for IAA and thereby it increases rooting. In avocado micro cuttings, IBA increased endogenous IAA and indole-3-acetyl-aspartic acid (IAAsp) concentrations before root differentiation and as the root formation proceeded (Hartmann *et al.*, 2002).

Solanki *et al.* (2012) studied the effect of different plant growth regulators on rooting in cuttings of Khair (*Acacia catechu*) by use of ten plant growth regulator treatments, three of IBA, three of NAA, three of IBA+NAA combination and control on cuttings. Among the different treatments of auxins used, treating the stem cuttings

with 500 mg L<sup>-1</sup> of IBA increased rooting per cent up to a maximum of 53.52 compared to 6.69 per cent in control. Other treatments like IBA 250 mg L<sup>-1</sup> + NAA 250 mg L<sup>-1</sup> (50.40 %) and IBA 250 mg L<sup>-1</sup> (40%) also helped in improving root per cent.

The cuttings of stevia treated with IBA 500 ppm were found to be superior with respect to sprouting percentage, shoot length, number of branches, number of leaves and root length and survival percentage. The study also revealed that better rooting and sprouting of stevia cuttings could be obtained through prolonged dipping of stevia cuttings in 500 ppm IBA solution (Chalapathi *et al.*, 1999)

Tomar and Singh (2011) studied the effect of air layering time with the aid of IBA in *Ficus krishnae* and *Ficus auriculata*. Maximum root formations of 80.25 per cent and 88.89 per cent were observed in pre-monsoon season in 500ppm IBA in *F. krishnae* and *F. auriculata* respectively. Rooting per cent increased with increased hormonal concentration in all three seasons (Monsoon, pre monsoon and post monsoon) in both species.

According to Reddy and Singh (1988), the non-wounded hardwood stem cuttings of guava treated with IBA 2000 ppm showed 87.5 per cent rooting and 62.86 per cent survival

Nautiyal *et al.* (2013) tried to increase the rooting of juvenile stem cuttings of *Quercus leucotrichophora* using chemical treatments and observed a significant variation in rooting percentage in all treatments using different concentration of IBA compared to control. Maximum rooting (36%) was discernible in the cuttings treated with 4000ppm, followed by 28 per cent in IBA 2000 ppm treated cuttings, whereas minimum rooting (24 %) was observed in untreated cuttings.

Amri *et al.* (2010) conducted a study on the effect of IBA on rooting success of *Diospyros melanoxylon*. Cuttings treated with 300 ppm of IBA gave 47.64 per cent rooting whereas control gave only 2.94 per cent rooting.

Majeed *et al.* (2009) reported that the cuttings of *Aesculus indica* was tested by using treatments with different concentrations of IAA, IBA and NAA. The cuttings treated with IBA 4000 ppm and 2000 ppm had a sprouting rate of 75 and 50 per cent, respectively, which was significantly higher than that of control and other treatments. All other treatments along with control failed to induce rooting. It was revealed that IBA 4000 ppm is a better-applied concentration for vegetative propagation of *A. indica* under Kashmir conditions.

The effect of auxins on macro propagation of *Taxus baccata* through cuttings was studied by Anjum *et al.* (2011). Application of IBA induced significantly higher

callusing percentage at 500 ppm (54.43%) as compared to control (2.70%). Application of IBA induced higher rooting percentage was achieved with 500 ppm of IBA (90%), which was significantly higher compared to other treatments whereas lowest rooting per cent was observed in control (1.92%).

Bagoury *et al.* (2006) studied the effect of IBA on survival percentage of stem cutting of stevia. Increase in IBA concentration from 0 ppm to 500 ppm and further to 1000 ppm increased the survival percentage from 74.44% to 82.44% and to 86.89%, respectively. Maximum value of survival percentage (95.33%) was obtained from basal cuttings treated with IBA at 1000 ppm while, the lowest value was associated with control (70.00%).

Camellia *et al.* (2009) applied different concentrations of IBA to semi hardwood and hardwood cuttings of *Jatropha curcus* to study its effect on rooting quality. Maximum rooting per cent was obtained in the treatment with 10000 ppm IBA for both semi hardwood (69%) and hardwood (79%) cuttings whereas control showed only 23 per cent and 30 per cent, respectively.

Henrique *et al.* (2006) applied different concentration of IBA, NAA and Paclobutrazol to *Pinus caribaea* cuttings to identify the best suitable treatment. The evaluation was performed 60 days after planting, and result showed that *Pinus caribaea* and *P.hondurensis* cuttings treated with IBA produce a higher percentage of rooted cuttings than those treated with NAA. The most effective treatment was IBA 4000mg L<sup>-1</sup> and 100mg L<sup>-1</sup> Paclobutrazol with 100 per cent rooting over 31 per cent in control.

Effect of auxins on adventitious rooting from stem cuttings of *Pongamia pinnata* was studied by Kesari and Krishnamachari (2008). All auxin treatments promoted sprouting and at lower concentrations triggered/enhanced rooting of cuttings. The effectiveness was in the order of IBA>NAA>IAA when applied singly. IBA at 4.92 mm was found to be most effective where rooting percentage and number of roots were significantly higher than in control.

Nair *et al.* (2005) reported successful rooting in vegetative propagation of *Melia dubia* through cuttings. Propagation using juvenile stem cuttings treated with 4000 ppm IBA showed promising result with 50 per cent rooting.

Husen and Pal (2007) studied effect of auxins treatment on rooting of teak cuttings with use of 2000 and 4000 ppm concentrations of IBA and NAA. In general, auxins treatment promoted overall quality of teak rooted cuttings. However, treatment with IBA increased percent rooting and percent sprouting while NAA suppressed it. The effect of each case was stronger when the higher concentration of each auxin was used.

Eganathan *et al.* (2000) compared the effect of different growth regulator on rooting of three mangrove species. Different concentrations of IBA, NAA and combination of IBA and NAA were applied to cuttings and layers of *Excoecaria agallocha*, *Heritiera fomes* and *Intsia bijuga*. The cuttings in all the three species responded better to IBA (2000-2500 ppm) over control, NAA or a combination of IBA and NAA. Maximum rooting was recorded when the cuttings and air layers were treated with IBA alone up to 2500ppm in all the three species.

Wendling *et al.* (2000) studied the effect of growth regulators on rooting frequency of eucalyptus stem cuttings by using various concentrations (0-6000 ppm) of IBA for root induction in cuttings of various clones of eucalyptus and found that 1000 ppm and 300 ppm dosages of IBA gives best result for maximum rooting in the cuttings of most of the clones.

Rooting was examined by applying various concentrations of IBA to the stem cuttings taken at different periods. One year old hardwoods stem cutting of three fig cultivars were planted in perlite from end of October to end of March. The highest rooting was obtained in the cv. Patlcan (58%) with 100 ppm IBA (Karadeniz, 2003).

The rooting studies on *Ficus mysorensis* stem cuttings by El-Hakim (1962), showed the stem cuttings treated with IBA 100 and 200 ppm solution for 12 hours showed highest rooting than IAA. Tilakaratna (1996) studied vegetative propagation in *Melia dubia* reported 89 per cent of rooting in *Melia dubia* cuttings without any treatment. Six month old seedlings were lopped at 10 cm below apical bud, and sprouts were taken from the plants after three days for the study. Sprouts planted in a highly shaded bed with river sand topping gave rooting without applying any growth regulator treatment.

Uppal and Khosla (1997) tried vegetative propagation through branch cuttings in selected temperate shrubs of Western Himalaya. IBA concentrations of 500 ppm, 1000 ppm, and 2000 ppm were used for vegetative propagation studies in *Dodonaea viscosa*, *Elaeagnus parviflora* and *Spiraea canescens*. A substantial increase in per cent success in rooting was noticed with application of IBA up to concentration of 1000 ppm in *S. canescens* and *E. parviflora* and 2000 ppm in *D. Viscosa*. *D. Viscosa* gave rooting per cent of 63.91 when treated with 2000 ppm IBA, whereas control gave only 48.83 per cent. In *S. canescens* and *E. parviflora*, cuttings treated with 1000 ppm IBA showed higher rooting per cent of 71.26 and 73.22, respectively when compared to the rooting per cent in control (48.08 and 45 %t).

Chhonkar and Singh (1972) reported that IBA at 5000 ppm was more effective than NAA in promoting the rooting and establishment of mango marcots. The

100 per cent rooting in mango marcots using NAA, IBA, NAA + IBA at 2500, 5000 and 10000 ppm and recorded 100 per cent survivability after transplanting.

In hardwood and semi hardwood stem cuttings of Karna Khatta (*Citrus kamaraj*) showed no significant differences observed by Singh (1962). But the better results were obtained with hardwood cuttings of vance (*Citrus liven*) reported by Singh and Singh (1973).

Mukherjee and Chatterjee (1978) confirmed that etiolated air layers of jack fruit (*Artocarpus heterophyllus* Lam.) treated with IBA at 10000 ppm and 5000 ppm gave 100 per cent and 91.66 per cent rooting, respectively. Further, they recorded a survival percentage of 91.66 and 74.75, respectively, after keeping in the nursery bed for one year. They also found that jackfruit air layers obtained from invigorated shoots and treated with 5000 ppm IBA gave 86 per cent rooting and 58 per cent survival after one year of planting in nursery bed.

In the studies conducted by Vijayakumar (1973) on propagation of guava by stem cuttings, 84.5 per cent rooting was obtained by treating the stem cuttings with 5000 ppm IBA and planting in the month of July. IBA treatment increased the number and length of primary roots. Stem cuttings of 'Allahabad Safeda' pretreated with 2000 ppm IBA + NAA + IBA, were found to have maximum rooting.

The studies done by Lenka and Das (1981) on litchi stem cuttings treated with 3000 ppm IBA transplanted in month of April in the medium perlite (0.5-1.0mm) showed highest rooting. Similarly, successful rooting in stem cuttings of mango cv. *longra* treated with 10000 ppm IBA.

Semi-hardwood stem cuttings of Kathbael (*Limonia acidissima*) immersed for 24 in 50 to 100 ppm NAA gave good results with respect to rooting and field. The studies on the rooting of *Allamanda cathodicab* soft wood cuttings treated with 200 ppm of IBA showed 92.50 per cent rooting compared to semi-hardwood cuttings treated with 3000 ppm IBA (Kempegowda *et al.* 2006).

Treating of hardwood cuttings of cinnamon (*Cinnamomum zeylanicum*) with 2500 ppm IBA, resulted in rooting of 45 per cent of cuttings. The application of 5000 ppm NAA induced 22 per cent rooting in semi hardwood cuttings. Gupta *et al.* 1989 reported a maximum survival of 90 per cent of rooted cuttings in *Lagastreemia lancasteri*, this rooted cuttings had been rooted by using 400 ppm IBA.

### 2.2.2 Effect of growth regulators and air layering on sprout and rooting percent on other species

Air layering is a simple propagation technique that was perfected in China more than 4,000 years ago to create offspring that are genetically identical to their parent plants. A method of propagating plants by wounding a stem or branch, applying a hormone to the wound, wrapping the stem or branch with damp sphagnum moss and polyethylene sheet to encourage root formation and finally removing the rooted stem or branch as an independent plant (Chandregowda and Shivaprasad (2008).

### 2.2.3 Effect of girdling and etiolation on rooting

Bhattacharjee and Balakrishna (1993) confirmed that etiolated air layers of jack fruit (*Artocarpus heterophyllus* Lam.) treated with IBA at 10000 ppm and 5000 ppm gave 100 per cent and 91.66 per cent rooting, respectively. Further, they recorded a survival percentage of 91.66 and 74.75, respectively, after keeping in the nursery bed for one year. They also found that jackfruit air layers obtained from invigorated shoots and treated with 5000 ppm IBA gave 86 per cent rooting and 58 per cent survival after one year of planting in nursery bed.

Girdling in relevance to propagation such as air layering is the process of completely removing a strip of bark (consisting of secondary phloem tissue) around a stem or branches. Girdling helps in the accumulation of photosynthesis such as CHO's and other metabolites. The accumulation of CHO's helps in the initiation of roots from phloem tissue or the bark tissue at the girdled site. Etiolation is the process of subjecting a stem of plant towards dark condition. Etiolation normally favors the rooting of cuttings. It is established that increased synthesis and accumulation of auxins takes place near the tissues which are exposed to dark condition. (Hartmann and Kester, 1986).

In avocado (*Persea americana*) stem cuttings were difficult to root. However, Frolich (1961) obtained good rooting by subjecting them to girdling and etiolation. Girdled and etiolated soft wood pecan (*Carya illinoensis*) stem cuttings produced more highly branched and more vigorous root system than only girdled stem cuttings. Hard and soft wood stem cuttings of pecan (*Carya illinpinensis*) did not initiate roots even after girdling (Taylor and Odom, 1970).

Nanda and Anand (1970) opined that relation between starch content and rooting in *Populus nigra* and stated that the delayed rooting shows high starch content and vigorous rooting shows low starch content.

#### 2.2.4 Effect of growth regulators on rooting in air layering method

Sen and Bose (1959) studied the effects of different growth substances on rooting of jack fruit air layers. IAA, IBA and NAA at 1000, 5000 and 10000 ppm were applied in lanolin paste at the time of ringing in the middle of May and June. A significant increase in the percentage of rooting was obtained due to all the three growth substances and IBA of 5000 ppm concentration and IAA and NAA at 10000 ppm resulted in better rooting.

The effect of IBA and NAA individually and with equal mixture of both in lanolin paste in the concentration range of 0 to 1000 ppm on sapodilla air-layers was tested by Tripathy *et al.* (2003). Good results were obtained when the mixture of IBA and NAA in equal proportion was applied compared to the application of individual growth substances in equivalent concentration. Rooting improved with increasing concentration and was highest (72.6 %) with the mixture at 1000 ppm. The macro rotting period was greatly reduced by this treatment. The shortest period was 107 days as compared to 5 to 6 months taken by air layers in the normal course. Further, they reported that IBA was better than NAA.

Kempanna *et al.* (1961) in a trial on propagation of *Glyricidia maculate* by air layering observed that mixtures of NAA and IBA was better for rooting rather than either of the growth substances applied

In cashew, Chhonkar and Singh (1972) observed that 88 per cent rooting of air layers treated with 75 ppm IBA. Average number of roots produced, length and diameter were superior in case of IBA treatment to IAA or control. They also obtained 84.6 per cent rooting in cashew air layers by treating with IBA at 300 ppm compared to 46.23 per cent in untreated marcots.

Sen and Chakravarthy (1972) reported that the best results can be obtained by treating the marcots of cashew with 500 ppm IBA. They obtained highest number of primary and secondary roots in cashew air layers due to treatment with combination of IBA 300 at ppm NAA at 200 ppm and 10 ppm of 2,4-D. According to Sengupta and Thakur (2001) the rooting of jackfruit air layers was maximum (90 %) when they were treated with 5000 ppm IBA + NAA.

The more number of primary roots (19.00) and secondary roots per layer (6.01) of jack fruit was recorded in combined treatment of IBA and NAA at 5000 ppm and less number (6.01 and 3.4 respectively) in control. Maximum length of primary roots (11.39 cm) was noticed in the jack fruit layers treated with both IBA + NAA at 5000 ppm and minimum length (3.41 cm) in the control treatment (Sengupta and Thakur, 2001).

Extensive root development which is important for success in vegetative propagation of cashew by air-layering was accomplished by several workers (Rajashekhara 2004). Air layers of cashew treated with IBA plus NAA each at 5000 ppm were found to survive better (Anon, 1996).

Alamgir and Momtaz (2005) observed that limited number of roots in cashew air layers was one of the important factors responsible for poor establishment of layers which may be further accentuated by rough handling of layers. The jack fruit layers when treated with 500 ppm IBA showed maximum success of (85 %) (Sengupta and Thakur, 2001) and minimum (54.22 %) in control treatment.

---

---

# **MATERIAL AND METHODS**

---

---

### III MATERIAL AND METHODS

The present investigation titled "**Studies on vegetative propagation techniques in *Diospyrosebenum*J. Koenig: An important endangered tree species**" was conducted at the College of Forestry, Ponnampet, Kodagu, Karnataka during 2017-2018. Detailed methodology adopted to conduct the experiment is described hereunder.

#### **3.1 Description of the study area**

The experiment was carried out during 2017-2018 at the nursery of College of Forestry Ponnampet, located in the hilly zone (Zone 9), Kodagu district of Karnataka state. Ponnampet situated at 12°08.579' N latitude; 75°56.317' E longitudes and at an altitude of 856 m above Mean Sea Level (MSL).

##### 3.1.1 Climate

The study area lies in tropical humid conditions. Mean monthly temperature in the study area varied from 22 °C to 28 °C during the study period with a mean annual rainfall of 2642.6 mm, most of which received between June, 2017 to November 2017.

#### **3.2 Experiment on vegetative propagation**

The study was carried out to know the efficacy of different types of vegetative propagation viz., air layering and branch cuttings for the propagation of *Diospyrosebenum* J. Koenig

##### 3.2.1 Air layering

Air layering is another alternative method is quite useful in plants, which are difficult to root by means of other vegetative propagation methods. The experiment was laid during the second week of November when atmospheric temperature and humidity were ideal for rooting. Healthy, vigorous superior and mature trees (15-17 year old) of *Diospyrosebenum* were selected and in each tree four branches of sizeable thickness were selected for the air layering. Healthy and vigorous one year old lateral branches with 2±2.5 cm thick and 30-45 cm length from the tips were selected and the bark was removed with sharp knife. Scraping was done to ensure the complete removal of phloem and cambium to avoid pre mature healing, later the following auxin treatment were given.

### **Auxin- (IBA treatment)**

Five IBA treatments 0 (control), 500 ppm, 1000ppm, 1500ppm and 2000ppm were applied with five replication in the experiment. The IBA concentrations were prepared by dissolving the appropriate amount of IBA in Ethanol and volume made up to 50 gram of talcum powder. The spagnum moss was tied to the each branches after the IBA treatment.

#### 3.2.2 Branch cuttings

Propagules were collected from healthy mother trees at College of Forestry, Ponnampet. Mother trees were pruned during the month of November to get the softwood, semi hardwood and hardwood cuttings required for the study.

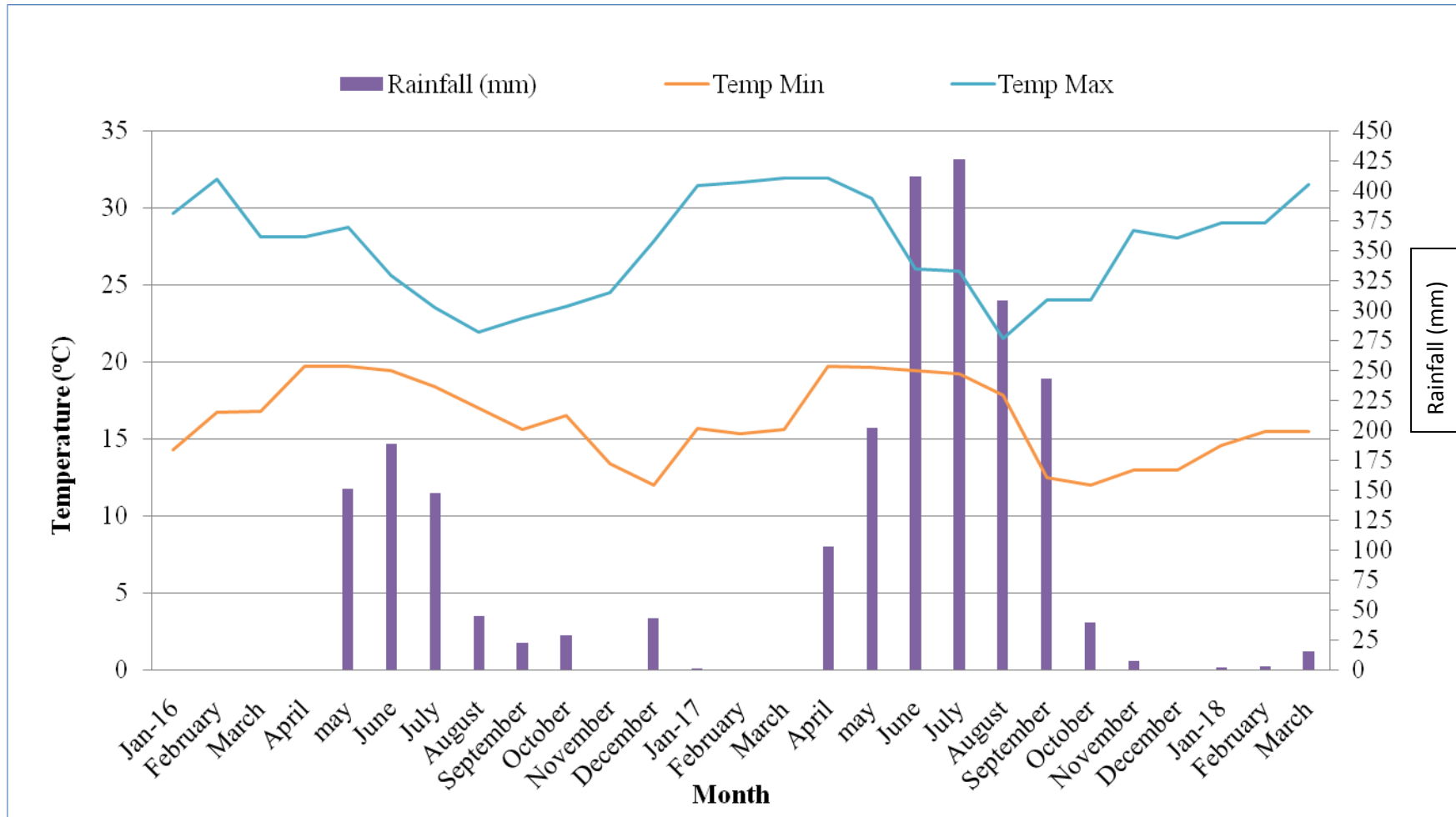
#### 3.2.3 Preparation of branch cuttings

Collected branches were brought to nursery for preparation of cuttings. Depending upon the formation of tissues, softwood cuttings, semi hard wood cuttings and hard wood cuttings with minimum of three nodes (10-15 cm length) were made from the collected shoots with secateurs. Prepared cuttings were treated with two percent carbendazim solution before application of growth regulator treatments.

Growth regulator used for treating the cuttings was Indole Butyric Acid (IBA). IBA was applied to the cuttings using dry dip smear method with three replication and the treated cuttings were kept in shade net for sprouting. The polythene bags were filled with standard nursery potting media soil sand and coir pith in the ratio 2:1:1. The details of treatment are as follows:

Treatment combinations followed during the study

Sl No.	Type of vegetative propagation	Growth regulator	Concentration (ppm)
1.	Air layering	IBA	Control
2.	Branch cuttings		500 (ppm)
			1000 (ppm)
			1500 (ppm)
			2000 (ppm)
<b>Based on success of rooting</b>			
<b>Type of cuttings</b>			
1.	Soft wood	IBA	Control
2.	Semi hardwood		500 (ppm)
3.	Hard wood		1000 (ppm)
			1500 (ppm)
			2000 (ppm)



**Figure 3.1. Meteorological observations during the study period from January 2016 to March 2018 at CoF, Ponnampet**

### 3.3 Data collection

The observation on primary data was collected at three intervals from November in the air layering and cutting which were treated with exogenous auxins.

#### 3.3.1 Observations for air layering

##### **Average number of roots**

Number of roots per area air-layered shoots was recorded by simple count method and average number of roots was calculated.

##### **Average length of roots**

Root length per air-layered shoots was measured by using 30 cm scale, and average root length was calculated.

##### **Rooting Percentage**

Rooting percentage was calculated by using the formula:

$$\text{Rooting percent} = \frac{\text{Number of air layered shoots rooted}}{\text{Total number of shoots air layered}} \times 100$$

#### 3.3.2 Observations for stem cuttings

Different growth parameters on which observations recorded at the end of the study (after 90 days of treatment) were as follows

1. Sprouting per cent =  $\frac{\text{Number of sprouted cuttings}}{\text{Number of cuttings planted}} \times 100$

2. Rooting per cent =  $\frac{\text{Number of rooted cuttings}}{\text{Number of cuttings planted}} \times 100$

3. Sprout length (cm) : The length of sprout was measured from the base of The sprout to the tip using scale and expressed in centimeter.

4. Number of leaves : Total number of leaves in the cutting was counted.

5. Rooting Success : Number of rooted cuttings to the total cuttings  $\times 100$

### 3.3.3 Biomass estimation

#### **Biomass of rooted cuttings**

The rooted cuttings from each treatment were marked to estimate the biomass at the end of the experiment. These cuttings were destructively sampled and washed in running water to remove adhering soil particles. Firstly, the total fresh weight was recorded using electronic balance and then, the rooted cuttings were cut at the collar region so as to divide them into root and shoot. Thereafter, fresh shoot weight and fresh root weight were recorded and expressed in grams.

#### **Estimation of dry weight**

Estimation of dry weight of the rooted cutting was done using the already cut shoot and root, which were separately dried at 70<sup>0</sup> C in paper bags for about 48 hours in hot air oven. Dry weight of shoot and root were recorded using electric balance and expressed in grams.

### **3.4 Statistical analysis**

The data collected from both the experiments were analyzed following proper procedures of data standardization for per cent values (Angular transformation) and count values (Square root transformation).

The interaction effects were subjected for two factor analysis to test the significant differences between vegetative propagation method and growth regulator used for the observations using the online statistical portal of CCS Haryana Agricultural University.



**Plate 3.1 Healthy selected *Diospyros ebenum* tree for the air layering**



**Plate 3.2 Different types of cuttings used for initiation of sprouts and roots in the study**

---

---

# **EXPERIMENTAL RESULTS**

---

---

## IV EXPERIMENTAL RESULTS

The present investigation titled "**Studies on vegetative propagation techniques in *Diospyros ebenum* J. Koenig: An important endangered tree species**" was conducted at the College of Forestry, Ponnampet, Kodagu, Karnataka during 2017-2018. The results of the investigations are presented here under in this chapter.

### **4.1 Initiation of new shoots by different vegetative propagation techniques and growth regulators**

The different vegetative propagation techniques *viz.*, air layering and branch cutting was adopted by selecting mature trees of *Diospyros ebenum* which were located in and around the campus. For the initiation of new shoots from the vegetative propagated method the different concentration of Indole Butyric Acid (IBA) was applied by dip smeared method. The observations on production of new shoots and root initiation in air layering and cuttings with hormonal treatment and their interaction studies were also taken.

#### 4.1.1 Root number, root length and leaf number in vegetative propagation techniques

The results of experiment treatment with different concentrations of IBA which were imposed to air layering and cuttings are presented in (Table 4.1). It was observed that in air layering the higher mean number of roots pr plants ( $6.16 \pm 1.98$ ) observed in treatment with 2000 ppm of IBA, followed by treatment with 1500 ppm IBA ( $5.51 \pm 1.47$ ). However, there was no production of roots and leaves in lower concentrations of IBA. The mean root length when followed through air layering with imposing 2000 ppm ( $1.59 \pm 0.40$ ) followed by 1500 ppm ( $1.36 \text{ cm} \pm 1.13$ ).

Production of root was also higher in treatment with IBA@ 2000 ppm ( $6.70 \pm 2.31$ ) and IBA of 1500 ppm ( $6.50 \pm 2.16$ ) through cuttings. However, the mean root length in cutting method was ( $1.73 \text{ cm} \pm 0.43$ ) with impose 2000 ppm followed by  $1.47 \text{ cm} \pm 0.23$  in treatment with 1500 ppm. The production of leaves in the air layering and cuttings were similar and it was observed that mean leaf number of  $2.5 \pm 1.29$  was noticed in treatment with 2000 ppm of IBA followed by 1500 ppm ( $1.5 \pm 1.29$ ).

### **4.2 Initiation of shoots and roots in types of cuttings**

Response of different concentrations of Indole Butyric Acid on rooting of cuttings was evaluated. The salient findings on effect of different concentrations of growth regulator on different vegetative propagation techniques were presented in Table 4.2.

**Table 4.1 Root number, root length and leaf number in different vegetative propagation techniques and concentration of Indole Butyric Acid**

Sl. no	Methods	Treatment	Root number (#) (Mean $\pm$ SD)	Root length (cm) (Mean $\pm$ SD)	Leaf number (#) (Mean $\pm$ SD)
1	Air layering	Control	0 $\pm$ 0	0 $\pm$ 0	0 $\pm$ 0
		500 ppm	0 $\pm$ 0	0 $\pm$ 0	0 $\pm$ 0
		1000 ppm	2.4 $\pm$ 2.81	0.22 $\pm$ 0.28	0.55 $\pm$ 0.95
		1500 ppm	5.51 $\pm$ 1.47	1.36 $\pm$ 1.13	1.5 $\pm$ 1.29
		2000 ppm	6.16 $\pm$ 1.98	1.59 $\pm$ 0.40	2.50 $\pm$ 1.29
2	Branch cuttings	Control	0 $\pm$ 0	0 $\pm$ 0	0 $\pm$ 0
		500 ppm	0 $\pm$ 0	0 $\pm$ 0	0 $\pm$ 0
		1000 ppm	2.95 $\pm$ 3.42	0.30 $\pm$ 0.35	0.75 $\pm$ 0.95
		1500 ppm	6.50 $\pm$ 2.16	1.47 $\pm$ 0.21	1.5 $\pm$ 1.29
		2000 ppm	6.70 $\pm$ 2.31	1.73 $\pm$ 0.43	2.5 $\pm$ 1.29

**Table 4.2 Response of cuttings to different concentrations of Indole Butyric Acid (IBA)**

SI No.	Types of Cuttings	Response	Concentrations of IBA (ppm)				
			Control	500	1000	1500	2000
1.	Soft wood cuttings	Number of new sprouts	0	0	2.5 ± 1.11	3.5 ± 0.50	3.75 ± 0.82
		Number of new leaves	0	0	2.13 ± 0.48	2.63 ± 0.48	3.13 ± 0.85
		Length of roots (cm)	0	0	0.59 ± 0.06	0.73 ± 0.18	1.36 ± 0.18
2.	Semi hardwood cuttings	Number of new sprouts	0	0	1.75 ± 0.82	4.5 ± 0.5	4 ± 0.70
		Number of new leaves	0	0	1.93 ± 0.36	2.18 ± 0.48	2.75 ± 0.63
		Length of roots (cm)	0	0	0	1.72 ± 0.36	2.32 ± 0.13
3.	Hardwood cuttings	Number of new sprouts	0	0	1.25 ± 0.43	2.00 ± 0.70	3.00 ± 0.70
		Number of new leaves	0	0	1.60 ± 0.24	1.96 ± 0.47	2.48 ± 0.48
		Length of roots (cm)	0	0	0	1.73 ± 0.14	2.17 ± 0.12

#### 4.2.1 Number of new sprouts

The effect of different concentrations of growth regulator on sprouting of *Diospyros ebenum* showed emergence of new sprouts except over control and 500 ppm of IBA concentration. Remarkably new sprouts were observed in all three types of cuttings as increase in concentration of IBA. The average higher number of new sprouts was observed in semi hardwood cuttings ( $4.5 \pm 0.50$ ) treated with 1500 ppm of IBA and 2000 ppm of IBA ( $4 \pm 0.70$ ). The least number of sprouts were recorded in hardwood cuttings ( $1.25 \pm 0.43$ ).

#### 4.2.2 Number of leaves

Observations on the number of leaves in rooted cuttings varied among the types of cuttings and concentration of IBA. Maximum number of leaves ( $3.13 \pm 0.85$ ) were observed in soft wood cuttings treated with T<sub>5</sub> (2000 ppm) and T<sub>4</sub> ( $2.63 \pm 0.48$ ). However, there was lesser number of leaves were noticed in hard wood cuttings of lower concentration of IBA 1000 ppm.

#### 4.2.3 Length of root (cm)

Effect of different concentrations of IBA on root length was evident in the present study Table 4.2 Higher root length ( $2.32 \text{ cm} \pm 0.13$ ) was recorded in semi hardwood cuttings followed by hardwood cuttings ( $2.17 \text{ cm} \pm 0.12$ ) treated with 2000 ppm of IBA. There was no initiation of roots in any of the cuttings treated with 500 ppm of IBA and control treatments.

### **4.3 Interaction effect of vegetative propagation method and IBA concentration on production of shoot and roots**

#### 4.3.1 Sprouting percent

There were non-significant differences were observed in sprouting percent, sprout length, production of leaves, rooting percent and rooting success in the main plot treatment air layering and cuttings. However, higher sprouting percent was recorded in air layering (5.55) as compared to cuttings (4.60) with a mean of (5.07).

There was a statistically significant differences observed in treatment concentrations of IBA. Sprouting per cent was found highest in T<sub>5</sub> (15.75) followed by T<sub>4</sub> (7.63). however, there was no sprouting occurred in T<sub>1</sub> and T<sub>2</sub> and lowest in T<sub>3</sub> (2.00).

Considering an interaction effect of vegetative propagation methods and IBA, there was no significant difference observed. However, interaction effect of M<sub>1</sub> × T<sub>5</sub> had higher sprouting per cent (17.50), followed by M<sub>2</sub> × T<sub>5</sub> (14.00) and least was recorded in M<sub>2</sub> × T<sub>3</sub> (1.75).

**Table 4.3 Effect of vegetative propagation methods, different IBA concentration and their interaction on growth attributes**

Sl. No.	Treatment	Sprouting (%)	Sprout length (cm)	No. of leaves (#)	Rooting (%)	Rooting success (%)
1	M <sub>1</sub> (Air layering)	5.55 (9.39)*	0.49	0.95 (1.33)#	3.69 (7.81)*	3.04
2	M <sub>2</sub> (cuttings)	4.60 (8.54)	0.45	0.95 (1.33)	4.46 (8.60)	3.65
3	Mean	5.07 (8.96)	0.47	0.95 (1.33)	4.07 (8.20)	3.34
	SEM±	0.59 (0.77)	0.03	0.18 (0.05)	0.34	0.21
	CD@5%	NS	NS	NS	NS	NS
1	T <sub>1</sub>	0 (0)	0	0 (1)	0 (0)	0
2	T <sub>2</sub>	0 (0)	0	0 (1)	0 (0)	0
3	T <sub>3</sub>	2.00 (5.74)	0.18	0.75 (1.28)	2.37 (6.28)	0
4	T <sub>4</sub>	7.63 (16.02)	0.72	1.50 (1.53)	8.76 (17.13)	7.25
5	T <sub>5</sub>	15.75 (23.07)	1.46	2.50 (1.84)	9.25 (17.64)	9.48
	Mean	5.07 (8.96)	0.47	0.95 (1.33)	4.07 (8.21)	3.34
	SEM±	0.94	0.05	0.28	0.55	0.33
	CD@5%	2.76	0.14	0.82	1.6	0.98
1	M <sub>1</sub> × T <sub>1</sub>	0 (0)	0	0 (1)	0 (0)	0
2	M <sub>1</sub> × T <sub>2</sub>	0 (0)	0	0 (1)	0 (0)	0
3	M <sub>1</sub> × T <sub>3</sub>	2.25 (6.11)	0.21	0.75 (1.28)	2.12 (5.94)	0
4	M <sub>1</sub> × T <sub>4</sub>	8 (16.42)	0.75	1.50 (1.53)	7.97 (16.33)	6.25
5	M <sub>1</sub> × T <sub>5</sub>	17.50 (24.43)	1.49	2.50 (1.84)	8.37 (16.79)	8.96
6	M <sub>2</sub> × T <sub>1</sub>	0 (0)	0	0 (1)	0 (0)	0
7	M <sub>2</sub> × T <sub>2</sub>	0 (0)	0	0 (1)	0 (0)	0
8	M <sub>2</sub> × T <sub>3</sub>	1.75 (5.37)	0.16	0.75 (1.28)	2.62 (6.61)	0
9	M <sub>2</sub> × T <sub>4</sub>	7.27 (15.63)	0.69	1.50 (1.53)	9.56 (17.93)	8.25
10	M <sub>2</sub> × T <sub>5</sub>	14.00 (21.72)	1.42	2.50 (1.84)	10.12(18.49)	10.00
	Mean	5.07 (8.96)	0.47	0.95 (1.33)	4.07 (8.20)	3.34
	SEM±	1.34	0.03	0.4	0.77	0.47
	CD@5%	NS	NS	NS	NS	NS

\* Values in parenthesis are angular transformed values

# Values in parenthesis are square root transformed values

T<sub>1</sub>: Control T<sub>2</sub>: 500 ppm T<sub>3</sub>: 1000 ppm T<sub>4</sub>: 1500 ppm T<sub>5</sub>: 2000 ppm

#### 4.3.2 Sprout length (cm)

There were non-significant differences observed for all the observations in main plot treatments. However, the maximum sprout length was observed in air layering (0.49 cm) as compared to branch cuttings (0.45 cm) with a mean of 0.47 cm.

Application of IBA concentrations showed statistically significant difference for sprout length. Highest sprout length was observed in T<sub>5</sub> (1.46 cm) and the lowest sprout length was found in T<sub>3</sub> (0.18 cm) with a mean of 0.47 cm.

Considering interaction effect of vegetative propagation methods and IBA, there was no significant difference observed for sprout length. Based on mean values, the interaction effect of M<sub>1</sub>×T<sub>5</sub> had higher sprout length (1.49 cm) followed by M<sub>2</sub>×T<sub>5</sub> (1.42) and least was recorded in M<sub>2</sub>×T<sub>3</sub> (0.16 cm).

#### 4.3.3 Number of leaves

There were non-significant differences observed for the average number of leaves among vegetative propagation types such as air layering and cuttings. However, overall mean number of leaves/cuttings in both type propagation techniques was 0.95.

There was a statistically significant difference observed in different concentrations of IBA. The number of leaves was found highest in T<sub>5</sub> (2.50) followed by T<sub>4</sub> (1.50). Lowest in T<sub>3</sub> (0.75). There was no leaf sprout in T<sub>1</sub> and T<sub>2</sub>.

Considering interaction effect of vegetative propagation methods and IBA concentration there was no significant difference observed for number of leaves per cuttings. However, the interaction effect of M<sub>1</sub>×T<sub>5</sub> and M<sub>2</sub>×T<sub>5</sub> had higher number of leaves (2.50) and least was recorded in M<sub>1</sub>×T<sub>3</sub> and M<sub>2</sub>×T<sub>3</sub> (0.75).

#### 4.3.4 Rooting (%)

There was non-significant difference observed for rooting per cent when air layering and cuttings considered. However, higher rooting per cent was observed in M<sub>2</sub> (cuttings) (4.46 %) compared to air layering (M<sub>1</sub>) (3.69 %) with a mean per cent rooting of 4.07.

There was a statistically significant difference among different treatments of concentrations of IBA. Higher rooting per cent was recorded in T<sub>5</sub> (2000 ppm) (9.25%) followed by T<sub>4</sub> (1500 ppm) (8.76%). The interaction effect of vegetative propagation methods and IBA, does not show significant difference for per cent rooting. However, based on mean values treatment of M<sub>2</sub>×T<sub>5</sub> achieved higher rooting

per cent (10.12%), followed by  $M_2 \times T_4$  (9.56%) and least was recorded in  $M_1 \times T_3$  (2.12%).

#### 4.3.5 Rooting success (%)

The highest rooting success was recorded in branch cuttings (3.65) as compared to air layering (3.04) with a mean of 3.34 per cent. There was a significant difference with respect to concentration of IBA. Among all the treatments, cuttings treated for rooting success with 2000 ppm of IBA ( $T_5$ ) recorded maximum rooting success (9.48%) followed by  $T_4$  (1500 ppm) (7.25%). The interaction effect of vegetative propagation methods and IBA, was found to be non significant mean value showed that treatment recorded for had higher rooting success  $M_2 \times T_5$  (10.00 %), followed by  $M_1 \times T_5$  (8.96 %) and it was least in  $M_1 \times T_4$  (6.25).

### **4.4 Effect of type of cuttings, concentration of IBA and its interaction on growth attributes**

The interaction effect of type of cuttings and IBA concentration on sprouting, sprout length, number of leaves, rooting per cent and rooting success was found to be non-significant and results are depicted in Table 4.4.

#### 4.4.1 Sprouting (%)

Even though there were non-significant differences observed with the type of cuttings, there was a higher sprouting percentage in semi hardwood cuttings (5.18 %) followed by softwood cuttings (4.25 %) with a mean of 4.55 per cent. When the concentrations of IBA considered, the highest sprouting percentage was noticed in  $T_4$  (6.17 %) followed by  $T_5$  (6.16 %). Interestingly, in all the concentrations there was sprouting but the least was recorded in control (2.85%) with a mean of 4.55%. However, the interaction effect of  $M_2 \times T_5$  had higher sprouting per cent (7.87%) followed by  $M_2 \times T_4$  (6.87 %) and least was recorded in  $M_3 \times T_1$  (2.56 %).

#### 4.4.2 Sprout length (cm)

The maximum sprout length was observed in softwood cuttings (0.53cm) followed by semi hardwood cuttings (0.46 cm) least sprout length was observed in  $M_3$  (0.43cm) with mean of 0.47 cm. Concentrations of IBA 1500 ppm ( $T_4$ ) showed higher sprout length (0.69cm) followed by 2000 ppm (0.58 cm) and the least sprout length was observed in  $T_1$  (control) (0.19 cm). The interaction effect of  $M_2 \times T_4$  had higher sprout length (0.68 cm) followed by  $M_3 \times T_4$  (0.63 cm) and least was recorded in  $M_3 \times T_1$  (0.14 cm).

**Table 4.4 Interaction effect of types of cuttings and IBA on production of shoots and roots**

SI No.	Treatment	Sprouting (%)	Sprout length (cm)	No. of leaves (#)	Rooting percent (%)	Rooting success (%)
1	M <sub>1</sub> (Softwood)	4.25 (9.18)*	0.53	1.57 (1.53)#	2.10 (5.23)*	3.65
2	M <sub>2</sub> (Semi hardwood)	5.18 (10.19)	0.46	1.37 (1.48)	1.87 (4.94)	3.33
3	M <sub>3</sub> (Hardwood)	4.22 (9.04)	0.43	1.21 (1.43)	1.52 (4.42)	3.13
	Mean	4.55 (9.47)	0.47	1.38 (0.10)	1.83 (4.86)	1.08
	SEM±	1.01	0.12	0.30	0.61	NS
	CD@5%	NS	NS	NS	NS	NS
1	T <sub>1</sub>	2.85 (6.85)	0.19	0.98 (1.36)	0.66 (2.33)	2.39
2	T <sub>2</sub>	3.43 (7.55)	0.40	0.99 (1.36)	1.04 (2.93)	2.29
3	T <sub>3</sub>	4.14 (8.35)	0.50	1.16 (1.40)	2.16 (5.95)	4.16
4	T <sub>4</sub>	6.17 (12.40)	0.69	2.04 (1.69)	2.54 (6.43)	4.02
5	T <sub>5</sub>	6.16 (12.19)	0.58	1.73 (1.60)	2.75 (6.67)	4.00
	Mean	4.55 (9.46)	0.47	1.38 (1.48)	1.83 (4.86)	3.37
	SEM±	1.31	0.15	0.39	0.79	1.40
	CD@5%	NS	NS	NS	NS	NS
1	M <sub>1</sub> × T <sub>1</sub>	3.00 (7.08)	0.23	1.12 (1.40)	0.75 (2.49)	2.62
2	M <sub>1</sub> × T <sub>2</sub>	3.50 (7.64)	0.45	1.00 (1.36)	1.25 (3.22)	2.50
3	M <sub>1</sub> × T <sub>3</sub>	3.87 (8.07)	0.55	1.50 (1.49)	2.62 (6.60)	4.37
4	M <sub>1</sub> × T <sub>4</sub>	6.12 (12.44)	0.76	2.37 (1.77)	2.87 (6.85)	4.50
5	M <sub>1</sub> × T <sub>5</sub>	4.75 (10.68)	0.66	1.87 (1.64)	3.00 (6.98)	4.25
6	M <sub>2</sub> × T <sub>1</sub>	3.00 (7.03)	0.18	1.00 (1.36)	0.75 (2.49)	2.43
7	M <sub>2</sub> × T <sub>2</sub>	3.67 (7.83)	0.38	1.06 (1.38)	1.00 (2.88)	2.25
8	M <sub>2</sub> × T <sub>3</sub>	4.50 (8.72)	0.50	1.06 (1.37)	2.12 (5.92)	4.12
9	M <sub>2</sub> × T <sub>4</sub>	6.87 (13.20)	0.68	2.00 (1.67)	2.62 (6.57)	3.87
10	M <sub>2</sub> × T <sub>5</sub>	7.87 (14.15)	0.56	1.75 (1.61)	2.87 (6.85)	4.00
11	M <sub>3</sub> × T <sub>1</sub>	2.56 (6.45)	0.14	0.83 (1.31)	0.50 (2.03)	2.12
12	M <sub>3</sub> × T <sub>2</sub>	3.12 (7.18)	0.37	0.92 (1.34)	0.87 (2.69)	2.12
13	M <sub>3</sub> × T <sub>3</sub>	4.07 (8.28)	0.47	0.93 (1.34)	1.75 (5.33)	4.00
14	M <sub>3</sub> × T <sub>4</sub>	5.51 (11.56)	0.63	1.77 (1.61)	2.12 (5.88)	3.68
15	M <sub>3</sub> × T <sub>5</sub>	5.87 (11.73)	0.52	1.58 (1.56)	2.37 (6.18)	3.75
	Mean	4.55 (9.46)	0.47	1.38 (1.48)	1.83 (4.86)	3.37
	SEM±	2.27	0.27	0.68	1.37	2.43
	CD@5%	NS	NS	NS	NS	NS

\* Values in parenthesis are angular transformed values # Values in parenthesis are square root transformed values

T<sub>1</sub>: Control T<sub>2</sub>: 500 ppm T<sub>3</sub>: 1000 ppm T<sub>4</sub>: 1500 ppm T<sub>5</sub>: 2000 ppm

#### 4.4.3 Number of leaves

The maximum number of leaves were recorded in soft wood cuttings ( $M_1$ ) (1.57) followed by  $M_2$  (1.37) with a mean of 1.38. When concentrations of IBA considered, the higher number of leaves were found in  $T_4$  (1500 ppm) (2.04) followed by  $T_5$  (1.73) and least was in  $T_1$  (control) (0.98). The interaction effect of  $M_1 \times T_4$  had higher number of leaves (2.37) followed by  $M_2 \times T_4$  (2.00) and least was recorded in  $M_3 \times T_1$  (0.83).

#### 4.4.4 Rooting (%)

$M_1$  (softwood) recorded maximum rooting (2.10 %) followed by  $M_2$  (1.87%) and least percent of rooting was occurred in  $M_3$  was 1.52% with a mean 1.83. with the application of 2000 ppm ( $T_5$ ) maximum rooting (2.75 %) was recorded followed by  $T_4$  (2.54 %) which was significantly superior over all other IBA concentrations. Interestingly rooting was observed in three treatments  $T_1$  (control),  $T_2$  (500 ppm),  $T_3$  (1000 ppm) were 0.66, 1.04 and 2.16 per cent, respectively. The interaction effect of  $M_1 \times T_4$  has higher rooting percent (4.50 %) followed by  $M_1 \times T_3$  (4.37 %) and least was recorded in  $M_3 \times T_1$  and  $M_3 \times T_2$  (2.12 %) respectively.

#### 4.4.5 Rooting success (%)

The highest rooting success was recorded in softwood cuttings (3.65) followed by  $M_2$  (semi hardwood cuttings) (3.33) and least was found in  $M_3$  (hardwood cuttings) (3.13) with the mean (3.34). Treatment with 1000 ppm of IBA ( $T_3$ ) recorded maximum rooting success (4.16) followed by  $T_4$  (1500 ppm) (4.02) and the least was in  $T_2$  (2.29). The interaction effect of  $M_1 \times T_4$  had higher rooting success (4.50) followed by  $M_1 \times T_3$  (4.37) and least was recorded in  $M_3 \times T_2$  (2.11).

### **4.5 Biomass of rooted cuttings under different concentrations of IBA**

#### 4.5.1 Softwood cuttings

##### 4.5.1.1 Fresh shoot weight (g)

Close perusal of the data presented in table 4.5 revealed that the fresh shoot weight for different IBA treatments varied from 0.30 g to 0.46 g. The estimated means of fresh shoot weight was found to be differing significantly among the different treatments. Significantly higher fresh shoot weight (0.46 g) was recorded by rooted cuttings in  $T_4$  (1500 ppm). Rooted cuttings of  $T_5$  and  $T_3$  showed fresh shoot weight of 0.43 g and 0.30 g, respectively and they were statistically on par with each other.

**Table 4.5 Biomass of soft wood cuttings treated with different concentration of IBA**

SI No.	Concentration of IBA	Fresh shoot weight (g)	Fresh root weight (g)	Total fresh weight (g)	Dry shoot weight (g)	Dry root weight (g)	Total dry weight (g)
1	T <sub>1</sub>	0.00	0.00	0.00	0.00	0.00	0.00
2	T <sub>2</sub>	0.00	0.00	0.00	0.00	0.00	0.00
3	T <sub>3</sub>	0.30	0.25	0.56	0.16	0.26	0.43
4	T <sub>4</sub>	0.46	0.36	0.83	0.28	0.20	0.49
5	T <sub>5</sub>	0.43	0.38	0.82	0.27	0.25	0.52
	Mean	0.23	0.19	0.44	0.14	0.14	0.28
	<b>SEM±</b>	<b>0.02</b>	<b>0.02</b>	<b>0.05</b>	<b>0.01</b>	<b>0.07</b>	<b>0.08</b>
	<b>CD@5%</b>	<b>0.07</b>	<b>0.08</b>	<b>0.15</b>	<b>0.04</b>	<b>NS</b>	<b>0.27</b>

T<sub>1</sub>: Control T<sub>2</sub>: 500 ppm T<sub>3</sub>: 1000 ppm T<sub>4</sub>: 1500 ppm T<sub>5</sub>: 2000 ppm

#### 4.5.1.2 Fresh root weight (g)

Estimated mean values of fresh root weight revealed the significant influence of IBA concentrations on fresh root weight. Among different treatments, the lowest value for fresh root weight was 0.25 g in 1000 ppm (T<sub>3</sub>), which was significantly inferior to fresh root weight found in T<sub>4</sub> (1500 ppm) which recorded 0.36 g and T<sub>5</sub> (2000 ppm) recorded maximum fresh root weight (0.38 g).

#### 4.5.1.3 Total fresh weight (g)

The data pertaining to total fresh weight showed a significant difference among different growth regulator treatments. Maximum fresh weight (0.83 g) was obtained in T<sub>4</sub> (1500 ppm), which was significantly superior to T<sub>5</sub> (2000 ppm) *i e*, 0.82 g. Least value of total fresh weight was obtained in T<sub>3</sub> (0.56 g).

#### 4.5.1.4 Dry shoot weight (g)

The data presented in Table 4.5 revealed that the dry shoot weight differed significantly among the treatments, consisting of growth regulator IBA. Maximum dry shoot weight of 0.28 g was obtained in T<sub>4</sub> (1500 ppm), followed by 0.27 g in T<sub>5</sub> (2000 ppm) and least value was 0.16 g in T<sub>3</sub> (1000 ppm).

#### 4.5.1.5 Dry root weight (g)

Estimated means of dry root weight did not differ significantly among the treatments of IBA concentration. However, value ranged from zero (lower concentration of IBA) to 0.26 g.

#### 4.5.1.6 Total dry weight (g)

The data presented in Table 4.5 shows the value of total dry weight as influenced by different growth regulator treatments. Total dry weight of rooted cuttings varied significantly among the treatments where, maximum total dry weight of 0.52 g was recorded in T<sub>5</sub> (2000 ppm), followed by 0.49 g in T<sub>4</sub> (2000 ppm). The least total dry weight of 0.43 g was recorded in 1000 ppm (T<sub>3</sub>). However, it was least in T<sub>3</sub> (1000 ppm) with 0.43g dry weight. Lower concentration of IBA does not influence on rooting.

### 4.5.2 Semi hardwood cuttings

#### 4.5.2.1 Fresh shoot weight (g)

Close perusal of the data presented in Table 4.6 revealed that the fresh shoot weight of semi hardwood cuttings for different IBA treatments varied from 0.44 g to 0.31 g. The estimated means of fresh shoot weight was found differ significantly

**Table 4.6 Biomass of semi hard wood cuttings treated with different concentration of IBA**

Sl. No	Concentration of IBA	Fresh shoot weight (g)	Fresh root weight (g)	Total fresh weight (g)	Dry shoot weight (g)	Dry root weight (g)	Total dry weight (g)
1	T <sub>1</sub>	0.00	0.00	0.00	0.00	0.00	0.00
2	T <sub>2</sub>	0.00	0.00	0.00	0.00	0.00	0.00
3	T <sub>3</sub>	0.31	0.24	0.55	0.14	0.12	0.28
4	T <sub>4</sub>	0.42	0.27	0.69	0.26	0.18	0.44
5	T <sub>5</sub>	0.41	0.35	0.77	0.24	0.23	0.48
	Mean	0.81	0.17	0.40	0.12	0.10	0.24
	<b>SEM±</b>	<b>0.02</b>	<b>0.02</b>	<b>0.04</b>	<b>0.16</b>	<b>0.03</b>	<b>0.04</b>
	<b>CD@5%</b>	<b>0.08</b>	<b>0.08</b>	<b>0.14</b>	<b>0.48</b>	<b>0.09</b>	<b>0.13</b>

T<sub>1</sub>: Control T<sub>2</sub>: 500 ppm T<sub>3</sub>: 1000 ppm T<sub>4</sub>: 1500 ppm T<sub>5</sub>: 2000 ppm

among the different treatments. Significantly higher fresh shoot weight (0.42 g) was recorded by rooted cuttings in T<sub>4</sub> (1500 ppm), which is followed by T<sub>5</sub> and T<sub>3</sub> with fresh shoot weight of 0.41 g and 0.31 g, respectively and were statistically on par with each other.

#### 4.5.2.2 Fresh root weight (g)

Estimated mean values of fresh root weight of semi hardwood cuttings revealed the significant influence of IBA concentrations on fresh root weight. Among different treatments, the lowest value for fresh root weight was 0.24 g in 1000 ppm (T<sub>3</sub>) which was significantly inferior to fresh root weight found in T<sub>5</sub> (2000 ppm) (0.35 g).

#### 4.5.2.3 Total fresh weight (g)

The data pertaining to total fresh weight of semi hardwood cuttings showed a very significant difference among different growth regulator treatments. Maximum total fresh weight (0.77 g) was obtained in T<sub>5</sub> (2000 ppm), which was significantly superior to total fresh weight recorded in T<sub>4</sub> (1500 ppm) *i.e.*, 0.69 g. Least value of total fresh weight was obtained in T<sub>3</sub> (0.55 g).

#### 4.5.2.4 Dry shoot weight (g)

The data presented in Table 4.6 revealed that the dry shoot weight of semi hardwood cuttings differed significantly among the treatments, with different concentration of IBA. Maximum dry shoot weight of 0.26 g was obtained in T<sub>4</sub> (1500 ppm), followed by 0.24 g in T<sub>5</sub> (2000 ppm) and least value was 0.14 g in T<sub>3</sub> (1000 ppm).

#### 4.5.2.5 Dry root weight (g)

Estimated means of dry root weight of semi hardwood cuttings differed significantly among the treatments. The highest of all (0.23 g) was found in T<sub>5</sub> (2000 ppm) followed by T<sub>4</sub> (1500 ppm) (0.18 g).

#### 4.5.2.6 Total dry weight (g)

The data presented in Table 4.5 shows that the total dry weight of semi hardwood cuttings recorded in different growth regulator treatments, varied significantly among the treatments of different concentration of IBA, maximum total dry weight of 0.48 g was recorded in T<sub>5</sub> (2000 ppm) followed by 0.44 g in T<sub>4</sub> (1500 ppm). The least total dry weight of 0.28 g was recorded in 1000 ppm (T<sub>3</sub>).

### 4.5.3 Hardwood cuttings

#### 4.5.3.1 Fresh shoot weight (g)

Close perusal of the data presented in Table 4.7 revealed that the fresh shoot weight of hardwood cuttings varied significantly from 0.25g to 0.36g, among various concentration of IBA treatments. Significantly higher fresh shoot weight (0.36 g) was recorded by rooted cuttings in T<sub>5</sub> (2000 ppm). Rooted cuttings of T<sub>4</sub> and T<sub>3</sub> were having fresh shoot weight of 0.33 g and 0.25 g, respectively and were statistically at par with each other.

#### 4.4.3.2 Fresh root weight (g)

Estimated mean values of fresh root weight of hardwood cuttings revealed the significant influence of IBA concentrations on fresh root weight. Among different treatments, the lowest value for fresh root weight was 0.20 g in 1000 ppm (T<sub>3</sub>) which was significantly inferior to T<sub>4</sub> (1500 ppm) which recorded 0.24 g. and T<sub>5</sub> (2000 ppm) 0.31 g.

#### 4.5.3.3 Total fresh weight (g)

The data pertaining to total fresh weight of hardwood cuttings showed a very significant difference among different growth regulator treatments (Table 4.7). Maximum fresh weight (0.67 g) was obtained in T<sub>5</sub> (2000 ppm), which was significantly superior to total fresh weight recorded in T<sub>4</sub> (1500 ppm) *ie*, 0.57 g. Least value of total fresh weight was obtained in T<sub>3</sub> (0.45 g).

#### 4.5.3.4 Dry shoot weight (g)

The data presented in Table 4.7 revealed that the dry shoot weight differed significantly among the treatments. Maximum dry shoot weight of 0.22 g was obtained in T<sub>4</sub> (1500 ppm), followed by 0.21 g in T<sub>5</sub> (2000 ppm) and least value was 0.12 g in T<sub>3</sub> (1000 ppm).

#### 4.5.3.5 Dry root weight (g)

Estimated means of dry root weight of hardwood cuttings differed significantly among the treatments of different concentration of IBA. The highest (0.22 g) dry root weight was found in T<sub>5</sub> (2000 ppm). T<sub>4</sub> (1500 ppm) recorded 0.16 g and T<sub>3</sub> (1000 ppm) recorded 0.10 g dry root weight of hardwood cuttings.

#### 4.5.3.6 Total dry weight (g)

The data presented in Table 4.7 shows that there was a significant difference for the total dry weight of hardwood cuttings different growth regulator treatments. Maximum total dry weight of 0.43 g was recorded in T<sub>5</sub> (2000 ppm) followed by 0.38 g in T<sub>4</sub> (1500 ppm). The least total dry weight of 0.23 g was recorded in 1000 ppm (T<sub>3</sub>).

**Table 4.7 Total fresh and dry weight (g) in hard wood cuttings with different concentration of IBA**

Sl No.	Concentration of IBA	Fresh shoot weight (g)	Fresh root weight (g)	Total fresh weight (g)	Dry shoot weight (g)	Dry root weight (g)	Total dry weight (g)
1	T1	0.00	0.00	0.00	0.00	0.00	0.00
2	T2	0.00	0.00	0.00	0.00	0.00	0.00
3	T3	0.25	0.20	0.45	0.12	0.10	0.23
4	T4	0.33	0.24	0.57	0.22	0.16	0.38
5	T5	0.36	0.31	0.67	0.21	0.22	0.43
	Mean	0.18	0.15	0.33	0.11	0.09	0.20
	<b>SEM±</b>	<b>0.02</b>	<b>0.02</b>	<b>0.04</b>	<b>0.01</b>	<b>0.03</b>	<b>0.04</b>
	<b>CD@5%</b>	<b>0.08</b>	<b>0.07</b>	<b>0.13</b>	<b>0.04</b>	<b>0.09</b>	<b>0.12</b>

T<sub>1</sub>: Control T<sub>2</sub>: 500 ppm T<sub>3</sub>: 1000 ppm T<sub>4</sub>: 1500 ppm T<sub>5</sub>: 2000 ppm

---

---

## **DISCUSSION**

---

---

## V DISCUSSION

Commercial exploitation of valued timber tree species hampered by the shortage of superior planting stock, primarily due to the difficulties experienced in propagating species using the conventional method of multiplication. *Diospyros ebenum* is a cross pollinated species exhibit a wide variability in terms of growth. Vegetative propagation techniques are used as indispensable tool for mass multiplication of superior phenotypes or genotypes and producing true to type uniform plants (Thakur *et al.*, 2008). Besides, vegetative propagation used for regeneration of tree species having problems of irregular seeding habits, long flowering and fruiting intervals, poor seed setting, low percentage of germination and undesirable short or long period of seed dormancy. Rooting of stem cuttings is easiest and economical methods of vegetative propagation usually exploited in many tree species. Only a limited success was achieved in rooting of stem cuttings of *Diospyros ebenum* species and concluded as difficult to root genus (Mewar and Naithani, 2016). Rooting of air layered and cuttings were affected by age of the ortet, season and exogenous application of root promoting hormones. It is essential to understand the critical factors in influencing the rooting (Siddiqui and Hussain, 2007). Therefore, an attempt was made to develop an efficient, economically viable and reproducible vegetative propagation protocol through air layering and branch cuttings for commercial propagation of selected trees of *Diospyros ebenum*: an endangered tree species.

The results of the present investigation titled “**Studies on vegetative propagation techniques in *Diospyros ebenum* J. Koenig: An important endangered tree species**” are discussed in the following sections.

### **5.1 New shoots and roots with different vegetative propagation method and growth regulators**

The vegetative propagation technique like air layering and branch cutting was adopted in selected mature trees of *Diospyros ebenum*. The different concentration of Indole Butyric Acid (IBA) was applied by dip smeared method to the air layered parts and branch cuttings. The present study revealed that IBA treatments were more efficient in inducing rooting of sprout and cuttings. The rooting percent steadily increased with an increase in concentrations of auxins from 500 ppm to 2000 ppm (plate 5.1). These results consistent and corroborated with the finding that IBA was most effective auxin in triggering rooting in air layering and cuttings (Jayaramkumar, 2006). It was observed that the higher number of roots per plant number (6.16) was recorded in treatment with 2000 ppm of IBA and there was no production of roots and leaves in lower concentrations of IBA. Similarly, production of root was also higher

in treatment with 2000 ppm (6.70). The mean root length (1.59) and leaf number (2.5) were also noticed with 2000 ppm of IBA. The auxin treatment significantly increased rooting percent, root length and number of leaves with exogenous applications of adequate IBA levels. The callus further differentiated into xylem leading to production of roots.

### **5.2 Shoots and roots in softwood cuttings**

The effect of different concentrations of growth regulator on shoot and root production in soft wood cuttings of *Diospyros ebenum* cuttings showed a significant difference and found maximum per cent of sprouting (3.75%), shoot length (2.48), number of leaves (3.13) and root length (1.36) in 2000 ppm of IBA as compared to other concentrations. This can be evident that at higher concentrations of auxins, there was vascular differentiation of cells and formation of roots which lead to enhanced formation of tissues. The study is also in line with Majeed *et al.* (2009) who reported that the cuttings of *Aesculus indica* treated with IBA @ 2000 ppm had a sprouting rate of 50 to 75 per cent.

### **5.3 Shoots and root in semi hardwood cuttings**

There was a higher sprouting of new shoots and shoot length as compared to soft wood cuttings of the species in semi hardwood cuttings (4.50) with 1500 ppm. But the higher shoot length, number of leaves and root length was recorded in IBA @ 2000 ppm. In semi hard wood cuttings, tissue sensitivity and release of chemicals lead to success of new shoots in 1500 ppm. These results were also in line with studies by Nair *et al.* (2005) where successful rooting of *Melia dubia* was recorded in juvenile stem cuttings treated with IBA @ 1500 ppm of IBA (50% rooting) and Camellia *et al.* (2009) applied different concentrations of IBA to semi hardwood and hardwood cuttings of *Jatropha curcus* and obtained maximum rooting (69 %) in 2000ppm.

### **5.4 Shoot and root in hardwood cuttings**

The hardwood cuttings treated with 500 ppm of IBA failed to produce adventitious shoots and roots. This is probably due to the presence of collenchymatous hypodermal tissues and vascular rays that are abutted against sclerenchymatous tissue. The fibrous hypodermal rings therefore act as a mechanical barrier for the emergence of roots Amri *et al.* (2010). Inability of rooting may also be due to insufficient carbohydrate content in stem cuttings, plant juvenility factors, C:N ratio of cuttings, age and positional effect of cuttings (Nanda and Anand 1970).

Rooting per cent as well as sprouting percent of cuttings was found to be dependent upon the concentration of growth regulator used for the study (Plate 5.2). The present study reported maximum per cent of rooting and sprouting with the use of



a) Control



b) 500 ppm



c) 1000 ppm



d) 1500 ppm



e) 2000 ppm

**Plate 5.1 Root and shoot initiation in different concentrations of growth regulator (IBA) in air layering**

higher concentration of growth regulator IBA @ 2000 ppm). Majeed *et al.* (2009) also support this finding with a similar study in *Aesculus indica*, which recorded maximum rooting and sprouting per cent with the use of maximum concentration of IBA (4000 ppm). Other than rooting and sprouting per cent, total stem height as well as collar diameter were also maximized with the use of higher concentration of IBA as found in this study.

During the study, length of root was found to be get effected by the concentration of growth regulator. A similar observation was also reported by Baul *et al.* (2010) in of *Litsea monopetala* where in the length of longest root was affected by growth regulator treatment. The present study reported the increase in the root length of branch cuttings as increase in concentration of IBA. Whereas the study of Baul *et al.* (2010) reported maximum root length in 0.1 per cent IBA, after which a negative impact was recorded along with increasing concentration.

### **5.5 Effect of IBA and vegetative propagation methods on shoot and root production**

The effect of interaction between IBA concentration, air layering and types of cuttings resulted in substanceous shoot and root production. Several authors documented rooting success of branch cuttings of different tropical forest trees (Leakey *et al.*, 2004; Husen and Pal, 2007; Baul *et al.*, 2010). IBA plays a significant role in rooting of various tropical forest tree species (Baul *et al.*, 2010). The use of IBA for rooting ability depends on the concentration of IBA among different species.

Sprout per cent was higher in air layering as compared to soft wood, semi hardwood and hardwood cuttings. This may be attributed to the hardy woody species of the nature of the tree produces more of phenolics in the cuttings lead to lower sprouting. Similarly, when higher concentration of 2000 ppm of IBA was applied to air layering, there was higher production of shoots and roots with increased sprout length and number of leaves. These findings also agree with the findings of Singh *et al.* (2003) in *Artocapus heterophyllus* cuttings with increased sprouts.

The lower ability of rootings in cuttings may be due to lower amount of endogenous auxin present in the cuttings of some species may reduce rooting ability (Baul *et al.*, 2010). At the same time, higher level of auxin content in the cutting of some species may lead to increase rooting ability without using of IBA or other growth hormones and postulated that higher concentration of IBA treatment may have some negative impacts against naturally occurring growth hormones in the cuttings.

## **5.6 Effect of IBA and vegetative propagation method on root initiation**

Higher concentrations of IBA in the study lead to increased rooting in cuttings used and the combination of cuttings and higher concentration of IBA also lead to increased root initiation, collar diameter, root length, number of roots and rooting success. Similar findings were also noticed by Ruiz *et al.* (2005) when evaluated the effects of IBA levels on rooting capacity of three type juvenile cuttings (apical, intermediate and basal) of *Gmelina arborea* and reported that the rooting and sprouting (54.9%) capacity than the basal ones (43.7 and 38.3% respectively) with 30% more root formed. IBA applications inhibited rooting capacity of apical cuttings, but stimulated it on interactions and basal cuttings the highest rooting percentage was obtained apical cuttings without IBA (80%) however, no significant differences in carbohydrates content were found among cutting types. Basak *et al.* (2003) tested the possibility of artificial regeneration through vegetative propagation of *Aegiceras corniculatum*. Rooting in air layer was induced with IAA, IBA and NAA. Maximum rooting ability was recorded in the air-layers treated with IBA 2500+NAA 5000 ppm with a promising rate of survivability.

## **5.7 Effect of IBA and softwood, semi hardwood and hardwood cuttings on production of shoots**

The hardwood cuttings of the species resulted in higher sprouting as compared to softwood and semi hardwood cuttings might be due to higher accumulation of starch content and reserved food substrates. However, there was gradual decrease in growth of sprouts and number of leaves in hardwood cuttings as compared to softwood and semi hardwood cuttings. This might be due to exhaust of reserve starch and food substrates and more cell elongation, tissues in soft wood and semi hardwood sprouts leads to increased growth and number of leaves. In the present study there was increased shoot growth and number of leaves in soft wood cuttings treated with IBA treatment of 1500 ppm alone Similar findings was also noticed by Kochhar *et al.*, (2005) where vegetative propagation of *Jatropha curcas* and *J. grandulifera* through stem cuttings and the accompanying biochemical changes. During the rooting process many changes took place both at the physiological and biochemical levels and activities of many enzymes were up and down regulated. Position of the cuttings on the mother branches also played a significant role in rooting and sprouting. Cuttings made from the middle portion of the mother branches exhibited better rooting compared to the most apical or most basal cuttings.



a) 1000 ppm



b) 1500 ppm



c) 2000 ppm

**Plate 5.2 Root and sprout initiation in different concentrations of growth regulator (IBA) in cuttings**

### **5.8 Effect of IBA and softwood, semi hardwood and hardwood cuttings on production of rooting**

The rooting initiation, development and growth of roots in treatment with different concentrations of IBA and types of cuttings used were varied. Maximum rooting was recorded in soft wood cuttings rather than semi hardwood and hard wood cuttings. IBA of 2000 ppm concentration yielded better rooting per cent, and root growth as well as success in rooting. But the regenerate of propagules were failed to establish in polythene bags filled with potting mixture in the mist chamber. This might be due to exhaust of reserve food molecules and accumulated starch in propagules. The similar findings were also noticed in studies by Gehlot *et al.* (2014) who reported that mini-cuttings of *Azadirachta indica* when treated with IBA of 1500 ppm had resulted in 80 per cent rooting. Similarly, Solanki *et al.* (2012) studied the effect of different plant growth regulators on rooting in cuttings of Khair (*Acacia catechu*) and IBA increased rooting per cent up to a maximum of 53.52 and Amri *et al.* (2010) reported that *Diospyros melanoxylon* cuttings treated with 300 ppm of IBA gave 47.64 per cent rooting.

### **5.9 Biomass of rooted cuttings under different concentrations of IBA**

The total dry weight of rooted propagules was influenced by the concentration of growth regulator used. In the present study the total dry weight of propagule was 0.50 g in air layering with 2000 ppm IBA, 0.52 gm in soft wood cuttings, 0.48 g in semi hard wood and 0.43 g in hard wood cuttings of *Diospyros ebenum*. These results similar to earlier report of Camellia *et al.* (2009) in the species *Jatropha curcas*. Among the different IBA concentrations used, maximum root weight was obtained with the highest concentration of IBA used (10000 ppm).

## **Conclusion**

- Among the vegetative propagation methods, semi hardwood cuttings was found to be suitable for *Diospyros ebenum*.
- Treatment with IBA 2000 ppm for both air layering and branch cuttings resulted in early initiating roots.
- Hardwood cuttings of the species found inability in producing either shoots or roots even when treated with IBA 2000 ppm.
- The sprouts produced through cuttings or air layering fails to establish with seedling
- An interaction of IBA 2000 ppm and semi hardwood cuttings produced new leaves, higher roots and shoots elongation.

## **Future line of work**

- Hardy species needs higher concentration of auxins
- Combined growth regulators need to be standardized
- Tissue culture techniques need to be worked out for large scale production of planting materials.

---

---

# **SUMMARY**

---

---

## VI SUMMARY

An attempt was made in the present study to develop an efficient, economically viable and reproducible vegetative propagation protocol through air layering and cuttings for commercial propagation of selected phenotypes of *Diospyros ebenum*. The vegetative propagation studies in *Diospyros ebenum* was carried out at nursery of College of Forestry, Ponnampet Kodagu Karnataka. The superior trees were identified in and around College campus and used for air layering and cutting experiments treated with application of different concentrations of Indole Butyric Acid. The summary of an experiment entitled “**Studies on vegetative propagation techniques in *Diospyros ebenum* J. Koenig: An important endangered tree species**” is presented as follows

- It was observed that the higher mean root number (6.16) observed in treatment with 2000 ppm of IBA followed by treatment with 1500 ppm IBA (5.51). Among all the treatments, treating with 2000 ppm of IBA (T<sub>5</sub>) recorded maximum rooting success (9.48) followed by T<sub>4</sub> (1500 ppm) (7.25).
- Significantly higher fresh shoot weight (0.46 g and 0.43 g) was recorded by rooted cuttings in T<sub>4</sub> (1500 ppm). And T<sub>5</sub> respectively. Similarly, highest root weight was also obtained by same treatment (i.e., 0.38g and 0.36g respectively in T<sub>5</sub> and T<sub>4</sub>).
- Maximum fresh weight (0.83 g) was obtained in T<sub>4</sub> (1500 ppm), which was significantly superior to total fresh weight recorded in T<sub>5</sub> (2000 ppm) i.e., 0.82g.
- Maximum dry shoot weight of 0.28 g was obtained in T<sub>4</sub> (1500 ppm), followed by 0.27 g in T<sub>5</sub> (2000 ppm) and least value was 0.16 g in T<sub>3</sub> (1000 ppm).
- Estimated means of dry root weight did not differed significantly among the treatments, due to the influence of growth regulator over initial fresh root weight.
- The highest of all (0.26 g) was found in T<sub>3</sub> (1000 ppm) followed by T<sub>5</sub> (2000 ppm) recorded 0.25 g and T<sub>4</sub> (1500 ppm) recorded 0.20 g were statistically on par with each other.
- Total dry weight of rooted cuttings varied significantly among the treatments due to the influence of growth regulator treatments on total fresh weight.

- Maximum total dry weight of 0.52 g was recorded in T<sub>5</sub> (2000 ppm) followed by 0.49 g in T<sub>4</sub> (2000 ppm).
- Significantly higher fresh shoot weight (0.42 g) was recorded by rooted cuttings in T<sub>4</sub> (1500 ppm).
- Rooted cuttings of T<sub>5</sub> and T<sub>3</sub> were having fresh shoot weight of 0.41 g and 0.31 g respectively and were statistically on par with each other.
- Estimated mean values of fresh root weight revealed the significant influence of IBA concentrations on fresh root weight.
- Among different treatments, the lowest value for fresh root weight was 0.24 g in 1000 ppm (T<sub>3</sub>) which was significantly inferior to fresh root weight found in T<sub>5</sub> (2000 ppm) (0.35).
- Maximum fresh weight (0.77 g) was obtained in T<sub>5</sub> (2000 ppm), which was significantly superior to total fresh weight recorded in T<sub>4</sub> (1500 ppm) ie, 0.69 g.
- Maximum dry shoot weight of 0.26 g was obtained in T<sub>4</sub> (1500 ppm), followed by 0.24 g in T<sub>5</sub> (2000 ppm) and least value was 0.14 g in T<sub>3</sub> (1000 ppm).
- Estimated means of dry root weight differed significantly among the treatments, due to the influence of growth regulator over initial fresh root weight.
- Total dry weight of rooted cuttings varied significantly among the treatments due to the influence of growth regulator treatments on total fresh weight.
- Maximum total dry weight of 0.48 g was recorded in T<sub>5</sub> (2000 ppm) followed by 0.44 g in T<sub>4</sub> (1500 ppm).
- Significantly higher fresh shoot weight (0.36 g) was recorded by rooted cuttings in T<sub>5</sub> (2000 ppm).
- Rooted cuttings of T<sub>4</sub> and T<sub>3</sub> were having fresh shoot weight of 0.33 g and 0.25 g respectively and were statistically on par with each other.
- Estimated mean values of fresh root weight revealed the significant influence of IBA concentrations on fresh root weight.

- Among different treatments, the lowest value for fresh root weight was 0.20 g in 1000 ppm (T<sub>3</sub>) which was significantly inferior to fresh root weight found in T<sub>4</sub> (1500 ppm) which recorded 0.24 g.
- Maximum fresh weight (0.67 g) was obtained in T<sub>5</sub> (2000 ppm), which was significantly superior to total fresh weight recorded in T<sub>4</sub> (1500 ppm) *i e*, 0.57g.
- Maximum dry shoot weight of 0.22 g was obtained in T<sub>4</sub> (1500 ppm) followed by 0.21 g in T<sub>5</sub> (2000 ppm) and least value was 0.12 g in T<sub>3</sub> (1000 ppm).
- Estimated means of dry root weight differed significantly among the treatments, due to the influence of growth regulator over initial fresh root weight.
- Total dry weight of rooted cuttings varied significantly among the treatments due to the influence of growth regulator treatments on total fresh weight.
- Maximum total dry weight of 0.43 g was recorded in T<sub>5</sub> (2000 ppm) followed by 0.38 g in T<sub>4</sub> (1500 ppm).

---

---

## **REFERENCES**

---

---

## VII REFERENCES

- AGNIHOTRI, K. AND ANSARI, S. A., 2000, Adventitious rhizogenesis in relation to seasonal variation, size of culm branch cutting and IAA treatment in bamboos. *Indian Forester*, **126**(9): 971-984.
- AHLAWAT, S. P. AND SINGH, U. V., 2000, Mass propagation of *Bambus abalcooa*, *B. nutans* and *B. tulda* through culm cuttings. *Arunachal Forest News*, **18**(1&2): 35-40.
- AHLAWAT, S.P., GUPTA, V. K., KUMAR, R.V. AND DUTTA, A. 2003, Vegetative propagation of *Acacia nilotica* (L.) ssp. *indica* through air layering. *Indian journal of Agroforestry*. **5**(1&2): 106-108.
- ALAMGIR, A.N.M. AND MOMTAZ AHAMED, 2005, Effect of growth regulators on root formation in stem cuttings of *Rauvol fiaserpentine* Benth and their propagule development. *Bangladesh J. Bot.*, **34**(1) :7-10.
- AMRI, E., LYARUU, H. V. M., NYOMORA, A. S. AND KANYEKA, Z. L., 2010, Vegetative propagation of African Blackwood (*Dalbergia melunoxylon* Guill. & Perr.): effects of age of donor plant, IBA treatment and cutting position on rooting ability of stem cuttings. *New Forests*, **39**:183-194.
- ANJUM Q., SHARMA L.K., GANIE S.A., RATHER M.M., RATHER H.A. Effect of auxines on macro propagation of *Taxus baccata* Linn. Through stem cuttings. *The Indian For*, 2011; **137**(12): 1382-1385.
- ANONYMOUS, 1996, International niles for seed testing. *Seed ScL and Tech*.
- BABITHA, M., RAO, P. S., MERU, E. AND SREENIVASULU, A. 2002, Effect of Auxins on rooting stem cuttings of *Givotia rottleriformis* Griff. Via hydroponit. *The Indian Forester*. **128**(3): 266-270.
- BAGOURY, E. L., OLFAT, H., ALLAM, A. I., ABDELGHANY, A. M. ANDATTIA, A. E., 2006, IBA application to stevia stem cutting: Its productivity and quality of plant and ratoon crops. *Egypt J. Agric. Res.* **84**(3): 299.
- BANDOPADHYAY, D. P., NATH, N., PANDEY, H. S. AND YADAV, L. P., 1982, Effect of growth regulators in propagation of *Carissa* species by stem cuttings. *Indian Agriculture*. **26**(1): 57-63.

- BANIK, R.L., 1984, Macro propagation of Bamboos by pre-rooted and pre-rhizomed branch cuttings, *Bano Biggyan Patrika* (Bangladesh Forest Research Institute, Chittagong), **13**: 67- 73.
- BASAK, U.C., MAHAPATRO, S.G. AND DAS, P. 2003, Vegetative propagation of *Aegiceras corniculatum*, a tree mangrove: biochemical and anatomical basis of adventitious rooting. *Indian Journal of Forestry*. **26**(2): 162-167.
- BAUL, A., SHARMA, L. K., GANIE. S. A, RATHER, M. M. ANDRATHER, H. A. H., 2010, Effect of auxins on macro propagation of *Taxus buccata* Linn. through stem cuttings. *The Indian Forester*, **137**(12): 1382-1384
- BHAGAT, B.K., JAIN, B.P. AND SINGH, C., 1998, Growth substances and survival of hardwood cuttings of Guava (*Psidium guajava* L.), *Journal of Research*, **10**: 121-123.
- BHATTACHARJEE, S.K. AND BALAKRISHNA, M., 1993, Response of root forming hormones on the regeneration of adventitious roots and the survival of rooted cuttings of different species of ornamentalshrubs. *Progressive Horticulture*, **25**: 13-17.
- BHOJVAID, P.P. AND NEGI, S. 2003, Propagation of *Elaeocarpus granitrus* by air layering. *The Indian Forester*.**129**(10): 1185-1191.
- BHUTANI, K.K., 2003, Effect of auxins on the rooting and survival of air layers in *Premna mucronata*. *Journal of Medicinal and Aromatic plant Sciences*. **25**(1): 63-65.
- BURDON, R.D. ANDAIMERS-HALLIDAY, J., 2003, Risk management for clonal forestry with *Pinus radiate* Analysis and review: Strategic issues and risk spread. *New Zealand Journal of Forestry Science*, **33**(2): 156–180.
- CAMELLIA. N. N. A., THOHIRAH, L A., ABDULLAH, N. A. P. AND KHIDIR, M.O., 2009, Improvement on rooting quality of *Jatropha curcas* using Indole Butyric Acid. *Research Journal of Agriculture and Biological Sciences*, **5**(4): 338-343.
- CHALAPATHI,M.V., THIMMEGOWDA, S.T., CHANDRAPRAKASH, J. ANDGANGADHAR, G. E., 1999, Vegetative propagation of Stevia (*Steviaria baudiana* Bertoni) under field conditions. *Crop Res.*, **18**: 319-320.
- CHANDRAMOULI, H., 2001, Influence of growth regulators on the rooting of different types of cuttings in *Bursera penicillata* (DC) Engl. *M.Sc.(Agri.) Thesis*, University of Agricultural Sciences, Bangalore.

- CHANDREGOWDA, M. AND SHIVAPRASAD, B. L., 2008, Effect of grow the regulators and methods of application on rooting of thyme (*Thymus vulgaris*) cuttings. *Mysore J. Agric. Sci.*, **42**(1): 9-14.
- CHAUHAN, S. K., KHAJURIA, H. N. AND GERA, M. K., 2003, A note on asexual propagation in neem and dek. *Journal of Research*, **40**(2): 215-216.
- CHHONKAR, V. S. AND SINGH. R., 1972, Effect of plant growth regulators on air layering in cashew nut. *Indian J. Hort.*, **24**: 26-29.
- CHMIEL, H., 1985, The effect of NAA, IBA and IAA auxins and their mixture on rooting of carnation cutting Cv Scania. *Acta Hort.*, **11**:162- 167.
- DAS, P., BASAK, U. C. AND DAS, A. B., 1997, Metabolic changes during rooting in pre-girdled stem cuttings and air-layers in *Heritiera*. *Botanical Bulletin of Academia Sinica*. **38**(2): 91-95.
- DAWSON AND WERE., 1997, Vegetative propagation techniques for tree species stem or root cuttings, layering and micro propagation cuttings. *Agroforestry Today*. **9** (2): 6-9.
- DE-CAPUA, J., 2005, The congo basin forest summit held February 4<sup>th</sup> and 5<sup>th</sup> in Brazzaville. Report on Congo Basin Washington, US. p. 11.
- DHUA, R. S. AND SEN, S. K., 1982, Root generation in lichi (*Litchi chinensis* Sonn.) stem cuttings with auxin and auxin synergists under intermittent mist. *South Indian Horticulture*. **30**(3): 188-190.
- EGANATHAN, P., RAO, C. AND ANAND, A., 2000, Vegetative propagation of three mangrove tree species by cuttings and air layering. *Wetlands Ecology and Management*. **8**:281-286.
- EL-HAKIM, S., 1962, Effect of some growth substances on the rooting of *Phyllanthus nivosus* V. *atropurpurea* and *Ficus mysorensis* Heyne cuttings. *Agricultural Research Review*, **40**(3):41-55.
- FAO,(Food and Agriculture Organization) 2015, Global forest resources assessment. Food and Agriculture organization. Main report: FAO Forestry paper. doi.http:// www.fao.org/3/a-i4808e.Pdf
- FINKEL, T. AND HOLBROOK, N. J., 2002, Oxidants, oxidative stress and the biology of aging. *Nature*, **408**:239-47.
- FROLICH, E.F, 1961, Etiolation and rooting of cuttings In: *Proceedings of the International Plant Propagates Society*, PP. 277-283.

- GEHLOT, A., GUPTA, R. K., TRIPATHI, A., ARYA, I. D. AND ARYA, S., 2014, Vegetative propagation of *Azadirachta indica*: Effect of auxin and rooting media on adventitious root induction in mini-cuttings. *Adv. For. Sci.*, **1**(1):1-9.
- GHOSH, S.K. AND BASU, R.M., 1974, Metabolic changes during regeneration of roots on cuttings. *Indian Journal of Experimental Biology*, **12**: 166-168.
- GONZALES, L. L., MARCOS, J., OUMIO, J. AND NICOLAS, S. U., 1991. Propagation of Kawayan-Kiling through split culm cuttings. *Canopy International*. **17**: 547-550.
- GUPTA, B. B., KUMAR, A. AND NEGI, D. S., 1989, Rooting response of branch cuttings of *Lagastreia lancasteri* L. *Indian Journal of Forestry*. **12**(3):210-214.
- HAISSING, B. E. AND DAVIES, F. T., 1984, Carbohydrate accumulation and participation in *Pinus banksiana* seedlings and seedling cuttings. *Physiologia plantarum*, **61**: 13-19.
- HARTMANN, H. T. AND KESTER, D. E., 1986, *Plant Propagation, Principles and Practices*. Prentice-hall Inc., Englewood Cliffs, New Jersey.
- HARTMANN, H. T., KESTER, D. E., DAVIES, F. T. AND GENEVE, R. L., 2002, *Plant Propagation, Principles and Practices*, 6th edition, Prentice Hall of India Pvt. Ltd., New Delhi, India.
- HASSAN, M., 1977, Studies on vegetative propagation of bamboos, *Bano Biggyan Patrica*, **6**(2): 64-71.
- HENRIQUE, A., CAMPINHOS, E.N., ONO, E. O. AND PINHO, S. Z., 2006, Effect of plant growth regulators in the rooting of pinus cuttings. *Brazilian Archives of Biology and Technology*. **49**(2): 189-196.
- HUSEN, A. AND PAL, M., 2007, Effect of branch position and auxin treatment on clonal propagation of *Tectona grandis* Linn. f. *New Forests*, **34**:223-233.
- IUCN (International Union for Conservation of Nature) (2010) Red List of Threatened Species™ <http://www.iucnredlist.org/> Accessed Nov 27<sup>th</sup> 2010.
- JAYARAMKUMAR, K., 2006, Effect of geographical variation on content of tannic acid, gallic acid, ethyl gallate in *Terminalia chebula* fruits. *Nat. Prod.*, **2**(3-4): 170-175.
- KARADENIZ., 2003, A study on some fruit characteristics and propagation of these by hardwood cuttings of local fig cultivars grown in ordu (Turkey), *Acta Horticulturae*, 107-112,

- KAROSHI, V. R. AND G.V. HEGDE (2000) Vegetative propagation of *Pongamia pinnata* (L). Pierre. Hitherto a neglected species. *The Indian Forester*, 128(3): 348-350.
- KEMPANNA, C., LINGARAJ, D.S., AND CHANDRASEKHARIAH, S.R. 1961 Propagation of *Gliricidia maculata*, by air layering with the aid of growth regulators, *Sci. and Cult*, **27**: 85-96.
- KEMPE GOWDA, K., GANESH AND MAHENDRAKAR, P., 2006, Efficacy of growth regulators for rooting and sprouting of long peper cuttings (*Piper longum* Linn.).*Journal of Plantation Crops*, **34**(3):650-651.
- KESARI, V. AND KRISHNAMACHARI, A., 2008, Effect of auxins on adventitious rooting from stem cuttings of candidate plus tree *Pongamia pinnata* L, a potential biodiesel plant. *Springer*, 1-6.
- KOCHHAR, S., KOCHHAR, V. K., SINGH, S. P., KATIYAR, R. S. AND PUSHPANGADAN, P., 2005, Differential rooting and sprouting behavior of two *Jatropha* species and associated physiological and biochemical changes. *Current Science*.**89**(6): 936-939.
- KRISHNAN, P. R., DIVAKARA, B. N., SELVAM, K. P. AND NEELANNAVAR, T. N. (2003).Propagation of *Simarouba glauca* Linn. by air layering with the aid of Indole Butyric Acid. *My forest*. **39**(1): 61-64.
- KUMAR, A. (2000). Clonal propagation of *Acacia mangium* through rooting of cuttings. *Annals of Forestry*, **8**(2): 250-252.
- LARSEN, F. E., AND GUSE, W. E. 1997. Propagating deciduous and evergreen shrubs, trees and vines with stem cuttings. A pacific north-west cooperative extension publication, Washington, USA, **p**. 10.
- LEAKEY, R. B., 2004, Physiology of vegetative reproduction. *Encyclopedia of Forest Sciences*. Australia. **p**. 114
- LEAKEY, R. R. B., J. F. MESEN, Z. TCHOUNDJEU, K. A. LONGMAN, J. MCP DICK, A. NEWTON, A. MATIN, J. GRACE, R. C. MUNRO AND P. N. MUTHOKA (1990). Low technology techniques for the vegetative propagation of tropical trees. *Commonwealth Forestry Review*, **69**(3): 247-257.
- LENKA, P. C. AND DAS, R. C., 1981, A note on the effect of rooting media on rooting of litchi stem cuts under mist chamber. *Orissa J. Hort*. pp 40-41.

- MAJEED, M., KHAN, M. A. AND MUGHAL. A. H. 2009, Vegetative propagation through stem cuttings treated with plant growth regulators. *Journal of Forestry Research.*, **20**(2): 171-173
- MEWAR, D. AND NAITHANI, D.C., 2016, physicochemical properties of jackfruit (*Artocarpus heterophyllus* Lam) seed flour and starch. *Science Asia, Plant Archives*, **16**(2), 959-962.
- MILLER, R., AND WIEDENHOEFT, A. 2002, Identification guide–Tropical woods: Guide to the identification of tropical woods controlled under the convention on international trade in endangered species of wild fauna and flora. *Handbook of USDA Forest Service, Animal, and Plant Health Inspection Service, Canada*, p. 23.
- MISHRA, V. K., CHAUKIYAL, S. P. AND PAL, M., 2002., Air layering trails in *Azadirachta indica* A. Juss. *The Indian Forester*. **128**(1): 70-74.
- MUKHERJEE, S.K. AND CHATTERJEE, B.K., 1982, Effect of etiolation and growth regulators on air layering of jackfruit (*Artocarpus heterophyllus* Lam.). *Indian. Hort.*, **35**: 1-4.
- MUKHIA, P. K., 2005, General guidelines for management of bamboo in Bhutan, Royal Government of Bhutan, Ministry of Agriculture, Department of Forestry, Forest Resources Development Division, Trimphu. p. 21.
- NAIR, K. N., MOHANAN, C. AND MATHEW, G., 2005, Plantation technology for selected indigenous trees in the Indian peninsula. *Bois et Forests des Tropiques*; **285**(3): 17- 23.
- NANDA, K. K. AND ANAND, V. K. (1970). Seasonal changes in auxin effects on rooting of stem cuttings of *Populus nigra* and its relationship with mobilization of starch. *Plant Physiology*. **23**: 99-107.
- NAUTTYAL, S., RAZVI, S., PRAKASH, R. AND NAUTIYAL, D. P., 2013, Effect of chemical treatment on rooting response of juvenile stem cuttings of *Quercus leuco trichoyhora* (bun oak). *The Indian Forester*, **139**(11): 1027-1029.
- NEUWINGER, 2000, Neuwinger african traditional medicine. a dictionary of plant use and applications, role of biotechnology. *Trends in Biotechnology*, **207**: 292-296.
- PATEL, B. M. AND AMIR, R. S., 1981, *In situ* soft wood grafting in mango. *South Ind. Hortic.*, **29**: 90-94.

- PRASAD. J., A. RABBANI AND R. A. RAM (1988). Rooting of hardwood cuttings of guava (*Psidium guajava*) L. through bottom heat. *Prop. Hortic.*, **20**: 20-23.
- RAJASHEKHARA, 2004, Standardization of vegetative propagation of stevia (*Stevia boudiana* Bertoni) through stem cuttings. *M.Sc.(Hort.) Thesis*, Submitted to University of Agricultural Sciences, Bengaluru, India.
- REDDY, K.M. AND SINGH, R. N.,1988, Efficacy of plastic-house in propagation of guava, (*Psidium guajava* L.), through hardwood cuttings. *Indian J. Agric. Sci.*, **58**: 81-82.
- RUIZ-GARCIA, R., VARGAS, H. J., ALCALA, C. M. AND MONTER, V. A. 2005, Effect of indol butyric Acid (IBA) and type of cuttings on rooting of *Gmelina arborea* Roxb, *Revista Fitotecnia Mexicana*. **28**(4):319-326.
- SCHEMERBECK, S. AND NAUDIYAL, T. H., 2018, Effect of species propagation on seed yield, *Indian Journal of Tropical Forest Science*. **7**(1): 23-27.
- SEETHALAKSHMI, K. K., VENKATESH, C.S AND SURENDRAN, T., 1983, Vegetative propagation of bamboos using growth promoting substances in *Bambus abalcooa* Roxb. *Indian Journal of Forestry*, **6**(2):98-103.
- SEN, P.K. AND BOSE, P.K., 1959, Effect of growth substances on rooting of jack fruit (*Artocarpus heterophyllus* Lam.) layers. *Indian Agric.*, **3**: 43-47.
- SEN, P.K. AND CHAKRAVARTY, S.P., 1972, Effect of growth regulators on air layering of cashew. 3<sup>rd</sup> Int. Symp. On Subtropical and Tropic. Hort. Soc., India.
- SENGUPTA, S. AND THAKUR, S., 2001, Studies on the effect of growth regulators on rooting of air-layers of jackfruits (*Artocarpus heterophyllus* Lam.).*Orissa J. Hort.* **29**(1):63-65.
- SIDDIQUI, M. I. AND HUSSAIN, S. A., 2007. Effect of indole butyric acid and types of cuttings on root initiation of *Ficus Hawaii*, *Sarhad J. Agri.*, **23**(4): 141-145.
- SINGH S. R. NARAYANASWAMY P, BANIK B. C. SHYAMALAMMA S, SIMON L. 2003. Development of RAPD-based SCAR marker related to fruit cracking in jackfruit (*Artocarpus heterophyllus* Lam.) *Crop Research* **42**(3): 151-156.
- SINGH, N.R. AND SRIVASTAVA, R.P., 1982, Studies on various factors involved in soft wood grafting in mango. *Prog. Hart.*, **14**: 117- 120.

- SINGH, P., BAJWA, M.S. AND SINGH, R., 1973 Propagation of ber, *Zizyphus mauritiana* Lam.): 1. Effect of rooting and IBA on the stooling. *Plant Sci.*, **5**: 137-139.
- SINGH, S., ANSARI, S. H. AND KUMAR P., 2002, Clonal propagation of *Bambus anutans* through culm and culm-branch cuttings. *The Indian Forester*, **128**(1):35-40.
- SOLANKI, V. K. KUKADIA., M.U., PATEL,S.R. AND TANDEL.M.B., 2012, Effect of different plant growth regulators on rooting in cuttings of Khair (*Acacia chundra*.)*Journal of Non-Timber Forest Products*, **19**(2):89-92.
- STRYDEM, D.K. AND HARTMANN, H.T., 1960, Effect of Indole Butyric Acid on respiration and nitrogen metabolism in Marianna plum soft wood cuttings. *Proc. Amer. Soc. Hort. Sci.*, **16**: 125-133.
- SURYANARAYANA AND VENKATESHWARA, 1982, Effect of growthregulators on air layering in mango. *Indian J. Hort.*, **42**: 30-36.
- TAYLOR, G.G. AND ODOM, R.E., 1970, Some biochemical compounds associated with rooting of *Caryail linoensis* stem cutting. *J. Amer Soc. Hort. Sci.*,**95**: 146-151.
- THAKUR, M., RANA, R. C., AND THAKUR, S., 2008, Physiochemical evaluation of *Termina liachebula* fruits. *J. Non-Timber Forest Prod.*,**15**: 37-42.
- THATOI, H., MISHRA, P.K., OUSEPH, A., MOHANTY, J.R. AND ACHARJYO, L.N.2001, Vegetative propagation by stem cuttings with auxins of four mangrove (and associate) species of Bhitakanika, *Indian Journal of Tropical Forest Science*.**13**(1): 223-227.
- TILAKARATNA, D. 1996. Vegetative propagation of lunumidella. In: proc. of the second annual forestry symposium on management and sustainable utilization of forest resources, Sri Lanka, pp. 65-68.
- TOMAR, A. AND SINGH, V. K., 2011, Effect of layering time (season) with the aid of Indole Butyric Acid in *Ficus krishnae* and *Ficus auriculata*. *The Indian Forester*, **137**(12): 1363-1365.
- TOPO, K. K. 1997,A study on medicinal & aromatic plants.*Botanica*.**27**:2-5.
- TRIPATHY, A.N., PANDEY, G. AND SHUKLA, P.K., 2003, Effect of auxins on rooting behavior of *Euphorbia pulcherrima* wild. *ProgressiveHorticulture*, **35**: 111-113.

- UPPAL. R. AND KHOSLA. P. K., 1997, Vegetative propagation through branch cuttings in selected temperate shrubs of Western Himalaya. *Ann. For.*, **5**(1): 80-83.
- VIJAYAKUMAR, N., 1973, Studies on the propagation of guava (*Psidium guajava* L.) by cuttings. *Ph.D. Thesis.*, Univ. Udaipur.
- WENDLING, I., XAVIER, A., GOMES, M., PIRES, E. AND ANRADE, H. B., 2000, Effect of growth regulator (IBA) on propagation of *Eucalyptus spp.* clones by micro cutting. *Revista Arovore*, **24**(2): 187-192.