

# Rooting Hormone and Substrate Effects on Mini-Cloned Mulberry (*Morus Indica*)

## ABSTRACT

Manipulation of propagation is a common practice in many commercial plant to obtain healthy and resistance plant for good yield. Many propagation techniques such as cutting, grafting, layering were evolved focusing on low budget production of high quality planting materials. An experiment was conducted to focus on standardizing a sound protocol for mini clonal propagation of *M. indica* variety V1. Among different concentrations of growth hormones evaluated, apical cuttings treated with IBA at 3000 ppm recorded significantly higher sprouting per cent (82.39 %), rooting per cent (63.27 %), survival per cent (68.80 %), root length (22.22 cm), shoot length (22.05 cm), number of roots (14.40) and number of leaves (18.30) followed by NAA at 4000 ppm. Besides that, various rooting medium used, Soil: Coir pith: FYM (1:1:1) registered superior in survival per cent (14.26 %), sprouting per cent (67.50 %), rooting per cent (38.77 %) and number of roots (12.45) whereas Soil: Sand: Vermicompost (1:1:1) registered maximum performance in shoot length (16.98 cm), root length (16.42 cm) and number of leaves (14.18). Hence it was clearly evident that IBA at 3000 ppm treated plants raised under Soil: Coir pith: FYM medium suitable for mass multiplication of *M. indica*.

*Keywords: Mulberry, Mini clone, Rooting hormone, Rooting media,*

## 1. INTRODUCTION

Mulberry (*Morus*) belongs to the family Moraceae. Morphologically mulberry is a fast growing, deciduous woody perennial tree, with deep rooting system. There are about 68 species reported in genus *Morus* and majority of them occur in Asia, especially in China (24 species) and Japan (19). Continental America is also rich in *Morus* species and poorly reported from Africa, Europe and Middle East, and absent in Australia (1). On the other hand in India, there are many species of *Morus* are present like *M. alba*, *M. indica*, *M. serrata* and *M. laevigata* are grow wild in the Himalayas. Several varieties have been introduced belonging to *M. multicaulis*, *M. nigra*, *M. sinensis* and *M. philippinensis*. Most of the mulberry varieties cultivated in India belongs to *M. indica* types. In the world out of 68 mulberry species have been reported (2) only 16 species performs well in terms of leaf, fruit and timber production (3). In India mulberry has been cultivated throughout the year especially in southern states namely Karnataka, Andhra Pradesh, Tamil Nadu and Telangana.

Mulberry has potential economic importance in developing countries in Asia, where sericulture is an important industry. The silkworm, *Bombyx mori* L., solely feeds on the leaf of mulberry therefore sustainability of sericulture heavily depends on the production of high-quality mulberry leaf. Mulberry is preferred due to its leaf yield, delicious fruit yield and some species have medicinal and ornamental properties and others are used due to their environmental adaptability (4). Mulberry is one of the most economically important tree crops

whereas Man's interest in mulberry cultivation originated with the growth of civilization and his fascination for quality fabric that led him search for silk.

Mulberry is generally propagated by sexual and asexual method. Sexual propagation is done through seeds and asexual propagation includes cutting, grafting and layering. Different methods of vegetative propagation are widely followed in different countries according to the environmental conditions and soil nature (5). Though propagation through stem cutting is easy, it has some restrictions viz., low rooting potential, less number of harvests per plant and long juvenile period. Additional problem involved in developing saplings in nursery is the maintenance and management cost for 3-6 months (6). There are several factors which can affect the rooting potential of stem cuttings include the nature of species, cultivar needs, source, condition of stock plant and pest and disease attack. Besides these growing conditions such as media, mist, bottom heat, use of hormones, fertilizer and supplemental lighting are also significantly influenced (7). In order to overcome these limitations, it is planned to establish an alternate method which mulberry can be propagated rapidly in a cost-effective way.

Mini clonal technology is a technique which helps in the production of large number of plants in short time with limited space. Mini clone cuttings showed enhanced rooting potential, rooting speed, and quality root system with reduced cost when compared to stem cuttings. Additionally, this system offer propagules with increased uniformity and reduced topophysis effect (8). There are many tree species has been successfully propagated by mini cutting technique viz., *Melia dubia*, *Casuarina sp.*, *Eucalyptus sp.*, *Tectona grandis* and *Dalbergia sissoo* reported by Parthiban *et al.* (9). When compare to the traditional stem-cutting method of propagation, mini-cuttings have many advantages leading to operational, technical, economic, environmental and quality benefits. The labour cost is reduced, due to elimination of labour intensive operations. Many agronomic practices like soil preparation, irrigation, fertilizer application, weeding, pest and disease management, cuttings transport etc. can be done with less manpower in smaller indoor areas at very lower costs, where the amount of chemicals used is also drastically reduced (10).

Thus various studies have been carried out in growth of mulberry using stem cuttings but only scanty of information is available on propagation of mini cuttings. With this backdrop, an attempt has been made to develop Mini clonal technology in mulberry cultivation with the following objectives, to determine the rooting hormone for clonal propagation of *M. indica* and optimize the suitable rooting medium for clonal propagation of *M. indica*.

## **2. MATERIAL AND METHODS**

### **2.1. Selection of mother plant**

Mulberry variety V1 is one among the popular varieties of India. It is developed by Central Sericultural Research and Training Institute in Mysore. Its leaves are large, thick, smooth, glossy dark green and ovate and claimed to be resistant against leaf spot and moderately resistant to leaf rust disease. It has high photosynthetic efficiency with an yield potential of 60 MT/ha/yr. It depends on the type of soil, climate and availability of water. Though it is ruling variety most of farmers much prefer V1 as foliage for silkworm. Owing to these advantages, it was decided to use V1 as mother source to harvest propagules for mini clonal propagation (11).

### **2.2. Preparation of stem cuttings**

Semi hard wood cuttings were excised from selected V1 mulberry variety. The cuttings were further trimmed to a length of approximately 15 cm with minimum three to four active buds. A slanting cut was given at the basal part of all cuttings and they were treated with 0.2 per cent Carbendazim 50% WP for 10 minutes. Treated cuttings were planted in nursery bed and each bed was accommodated with 90 cuttings (12).

### **2.3. Nursery bed preparation**

Before commencement of experiment, the field was ploughed to make the soil fine and suitable for establishing mother garden. Two beds of 2m × 1m size were prepared and each bed was enriched with 1kg of FYM, 500g of *Trichoderma viride* and 500g of *Pseudomonas fluorescence* (13).

#### 2.4. Preparation of mini cuttings

Apical shoot cuttings were excised from mother garden and the cuttings were further trimmed to mini size. The mini-cuttings were harvested using sterile pruning scissors during early morning hours. All leaves in the shoot, except two leaves near the apical point were removed before using it for different hormonal treatments. A 45° angle slanting cut was made at the base of all the apical cuttings. After preparation, cuttings were given a prophylactic treatment against fungal disease using aqueous solution of 0.2 per cent Carbendazim 50 % WP for 20 minutes, subsequently washed with distilled water. After fungicidal treatment mini cuttings were subjected to various auxin treatments such as IBA and NAA (14). Further, the apical shoot cuttings were also dipped in 0.2 per cent Dichlorovos solution 76 % EC to control the leaf webber for 2 minutes.

#### 2.5. Rooting hormone and its concentration for growth of mini cuttings

Auxin is one of the major endogenous hormones known to be intimately involved in the process of adventitious rooting (15). The prepared apical shoot cuttings were treated with various rooting hormones such as Indole Butyric Acid and Naphthalene Acetic Acid powder for 15 minutes. Different concentration of rooting hormones viz., 1000, 2000, 3000, 4000 and 5000 ppm were used. The preparation of rooting hormone requires IBA and NAA powder, Boric acid crystal, Bavistin powder and talc powder. For 1000, 2000, 3000, 4000, 5000 ppm of selected concentration needs 0.1g, 0.2g, 0.3g, 0.4g, 0.5g of hormone using 100g of talc based formation. Selected Mini-cuttings were dipped in the rooting hormone and planted in polybags and raised under polytunnel. Observations on growth attributes viz., shoot length, root length, number of leaves and numbers of roots were taken at 90 DAP.

#### 2.6. Optimize the rooting medium for mini clonal propagation

After auxin treatment the apical shoot cuttings were planted in polybags containing different rooting medium. The 1/3 basal cut portion was inserted in different rooting media using sticking technique (16). In order to make complete wetting on entire cut surface, holes were made in each bag to avoid the damage of cambium and to prevent the removal of rooting hormone. Later the cuttings were planted in rooting medium are kept inside the low cost poly tunnels under shade net. Irrigation was done once in a week in order to maintain desired humidity of 80 to 90 per cent. The temperature in the poly tunnel was maintained at 33 ± 1°C. The studies were conducted in Factorial Completely Randomized Design (FCRD) with four replications, with 15 cuttings per replication. Most of the cuttings started rooting on 15 days after planting. The detailed accounts of treatments were given below

Chart 1 : The Treatment details.

Treatment	Rooting hormone	Concentration
T <sub>1</sub>	IBA	0, 1000, 2000, 3000, 4000, 5000 ppm
T <sub>2</sub>	NAA	0, 1000, 2000, 3000, 4000, 5000 ppm
	Rooting Media Mixture	Ratio
M1	Soil	-
M2	Soil+ Sand+ FYM	1:1:1
M3	Soil+ Coir pith	1:1
M4	Soil+ Sand+ Vermicompost	1:1:1
M5	Soil+ Coir pith+ FYM	1:1:1

## 2.7. Observations recorded

### Sprouting percentage

$$\text{Sprouting percentage} = \frac{\text{Number of cuttings sprouted}}{\text{Number of cuttings planted}} \times 100$$

### Rooting per cent

$$\text{Rooting percentage} = \frac{\text{Number of rooted cuttings}}{\text{Number of cuttings planted}} \times 100$$

### Survival percentage

$$\text{Survival percentage} = \frac{\text{Number of cuttings survived}}{\text{Number of cuttings planted}} \times 100$$

### Root length

The rooted cuttings were washed in water to remove the medium without disturbing the root system. The root length of each plant was initially measured from the terminal end of the shoot to the tip of the root using standard scale and expressed in cm.

### Number of roots per cutting

Number of roots was calculated by counting lateral roots present in a mini shoot.

### Shoot length

The shoot length was measured from the point of shoot initiation to tip of the shoot and expressed in cm.

### Number of leaves per cutting

Total number of leaves produced by mini cuttings was counted numerically.

## 2.8. Statistical design

All data were subjected to statistical analysis to assess the possible relationship between different parameters and analysis of variance was employed in nursery by adopting Completely Randomized Design and Factorial Completely Randomized Design. The stage wise data were analyzed separately using AGRES software.

## 3. RESULTS AND DISCUSSION

Auxin is one of the major endogenous hormones involved in the process of adventitious rooting (15) and the physiological stages of rooting are associated with changes in auxin concentration (17). Adventitious root formation in stem cuttings is a crucial physiological process for clonal propagation of many plant species. In the present study IBA @ 3000 ppm reigned best in growth traits viz., sprouting percentage (82.39 %), survival percentage (68.80 %), rooting percentage (63.27 %), number of leaves (18.30), number of roots (14.40), root length (22.22 cm) and shoot length (22.05 cm) (Fig.1). Usually exogenous application of auxins viz., IBA and NAA are recommended for promoting adventitious roots in stem cutting propagation of many shrubs (18) and trees (10). Current study revealed that among two different rooting hormones (IBA and NAA) used, IBA concentration recorded better survival percentage. IBA @ 3000 ppm noted to be the highest survival per cent (68.80 %) followed by NAA @ 4000 ppm (57.72 %) (Fig.3). These results are agreed with Rahdari *et al.* (19) in *Aralia* semi-wood cuttings. High survival per cent is considered due to care requirement in propagation unit.

Koyuncu and Senel (20) found the highest survival per cent (80.00 %) in black mulberry. In addition to that, IBA @ 3000 ppm registered the maximum survival per cent (74.33 %) in *Psidium guajava* cuttings reported by Rani *et al.* (21). The increased survival per cent is due

to increase in number and length of root due to effective intake of nutrients and water (22). Similar studies were reported by Abdullah *et al.* (23) and Sukhjit (24) in hardwood cuttings of guava and peach. In the present investigation IBA @ 3000 ppm showed the highest sprouting of 82.39 per cent followed by NAA @ 4000 ppm was 68.44 per cent and IBA @ 2000 ppm was 66.35 per cent. Similar to present findings of Singh *et al.* (25) mulberry cuttings treated with IBA @ 2000 ppm are obtained maximum number of sprouted cuttings (9.67) followed by NAA @ 2000 ppm (5.67). This might be due to better root growth, absorption and translocation of nutrients to plant parts (26). In addition to that, Husen (27); Husen (28) reported similar findings in *Tectona grandis*.

In general hormone application promotes and increases the root formation. In the present study different concentrations of IBA and NAA were used, IBA at 3000 ppm recorded the highest rooting per cent compared to other treatments. Rooting in IBA @ 3000 ppm was 74.00 per cent followed by NAA @ 4000 ppm was 39.00 per cent (Table.1, Fig.2). Kalyoncu *et al.* (29) observed the highest rooting per cent (100 %) was obtained from the application of IBA at 3000 ppm in black mulberry cuttings. This findings were also supported by Koyuncu and Senel (20) who reported that black mulberry cuttings treated with 5 g/lit IBA in bunch planting method suited for its better rooting. The increase in auxin concentrations led to increase in rooting per cent in oleander plant up to 3000 ppm IBA and subsequent increase in IBA leads to decrease in rooting reported by Habibi *et al.* (30). A higher dose of auxin (200 ug per cutting) shows inhibition in rooting of certain clones of *Triphlocliton scleroxylon* cuttings (31).

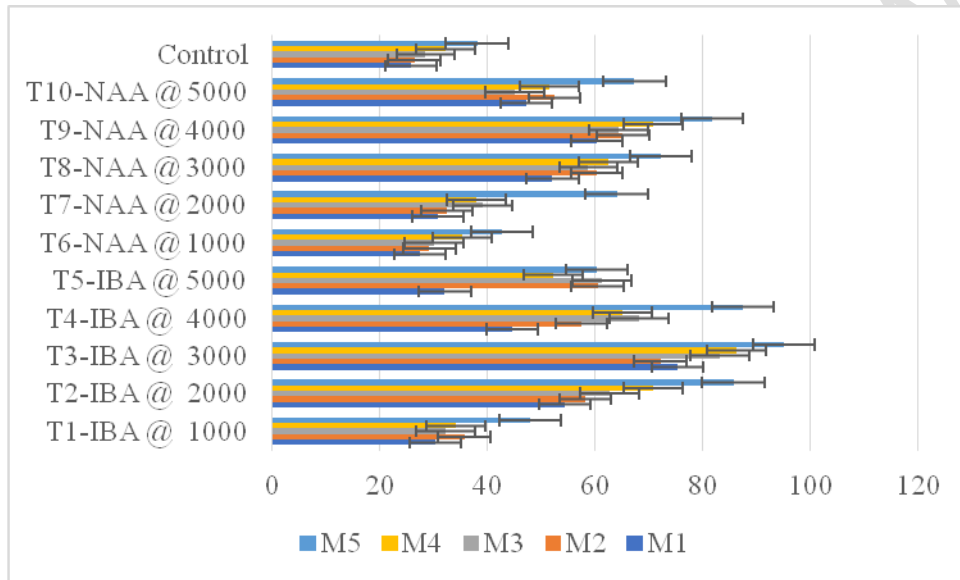
Similarly, when auxin concentration increases beyond 3000 ppm rooting was found to be decreased. On contrary to this study auxin application increased the rooting per cent in tree species (24; (32) in *Bougainvillea glabra*; (33) in *Citrus lemon* cv. Cuttings; (34) in teak mini cuttings). The study reported that maximum number of roots recorded in IBA treated plants compared to NAA. IBA @ 3000 ppm registered maximum number of roots (14.40) at 90 DAP followed by NAA @ 4000 ppm (12.45)(Table.2). These results are in accordance to findings of Rani *et al.* (2018) who reported that apical cuttings treated with IBA (3000 ppm) produced more number of roots in *Psidium guajava* and this could be due to accumulation of essential internal substances and facilitates downward movement of nutrients to roots.

The present investigation is in consonance with Ullah *et al.* (35) who stated that induction of more roots in cuttings due to stimulation of cambium activity by hormonal application in many species. The variation in dose response to the number of roots might also be attributed to the varietal and climatic differences in the location (36). Among all treatments IBA recorded maximum root length. At 90 DAP the maximum root length registered in IBA at 3000 ppm was 22.22 cm followed by NAA at 4000 ppm (20.62 cm). Similar observations were made by Singh (37) in mulberry reported that maximum root length was influenced by hormonal application. According to Ghatnatti (38) maximum root length was possible to the action of auxin activity due to hydrolysis and translocations of carbohydrates towards base of cuttings are lead to cell division and elongation. This result falls in line with Baroudi *et al.* (39) who reported that softwood cuttings of mulberry (*M. alba*) treated with IBA (2000 ppm) produced good root length, more number of roots and rooting per cent. The present findings are also similar to the observations of Galavi *et al.* (40) in *Vitis vinifera* and Kumar (41) in *Melia dubia*.

In the present investigation, at 90 DAP, IBA at 3000 ppm recorded the maximum number of leaves (18.30) followed by NAA at 4000 ppm (12.05) (Table.3). This result are in line with findings of Pallavi *et al.* (36) who recorded the increase in number of leaves, more number of roots, plant height and more number of branches at 2000 ppm of IBA in mulberry cuttings. Similarly the positive influence of IBA on number of leaves might be due to activation of shoot growth led to an increased number of active nodes and similar conclusions were also given by Wahab *et al.* (42) in *Psidium guajava* L. cuttings, Siddiqui and Hussain (43) in *Ficus Hawaii* cuttings, Alam *et al.* (44) in Kiwi cuttings which supports the current study.

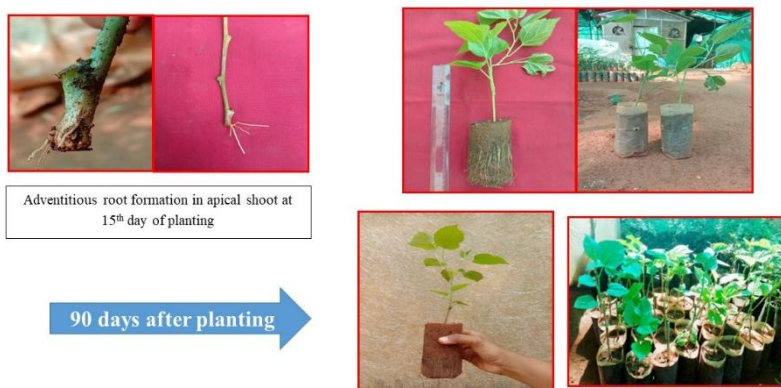
The maximum shoot length of 22.05 recorded in IBA at 3000 ppm at 90 DAP followed by NAA at 4000 ppm (21.51 cm). This increase in shoot length may be due to geotropism effect. These results are in agreement with Kiruthika (14) who found that *M. sinensis* mini cuttings treated with IBA at 5000 ppm showed the maximum shoot length (22.65 cm) at 90 DAP. In addition to that similar findings were also reported in *D. sissoo* (45); *Tectona grandis* (46) and other plant species.

**Fig 1. Interaction of rooting hormone and rooting medium on sprouting per cent of *M. indica***



M1- Soil M2- Soil: Sand: FYM (1:1:1) M3- Soil: Coir pith (1:1) M4- Soil: Sand: Vermicompost (1:1:1) M5- Soil: Coir pith: FYM (1:1:1)

**Fig 2. Root formation in mini clone cuttings in mulberry**

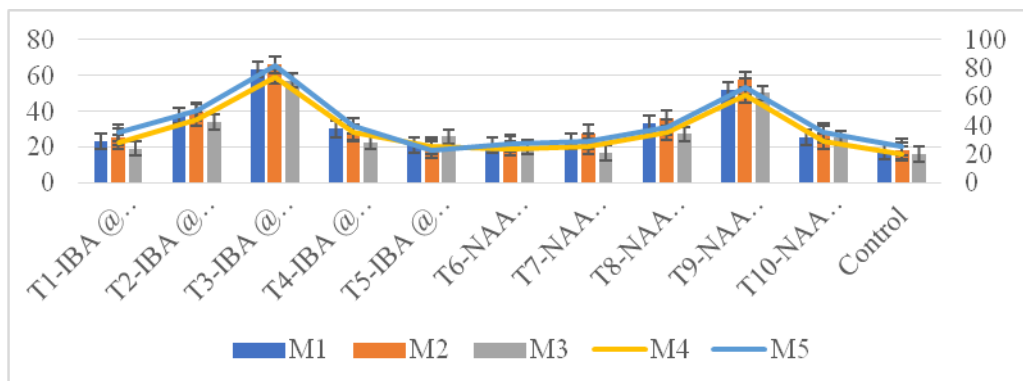


**Table 1. Effect of rooting hormone and rooting medium on rooting per cent of *M. indica***

Treatments	Shoot Length						Number of roots					
	M1	M2	M3	M4	M5	Mean	M1	M2	M3	M4	M5	Mean
IBA @ 1000	11.63	12.72	10.70	14.05	13.11	<b>12.44</b>	8.25	9.00	8.00	10.00	11.50	<b>9.35</b>
IBA @ 2000	15.03	16.00	14.23	18.37	16.22	<b>15.97</b>	11.00	12.25	10.50	13.50	14.25	<b>12.30</b>
IBA @ 3000	19.77	20.08	18.60	26.11	25.70	<b>22.05</b>	13.25	14.50	12.50	15.25	16.50	<b>14.40</b>
IBA @ 4000	13.82	14.73	12.20	16.81	15.81	<b>14.67</b>	10.50	12.00	9.25	13.00	13.25	<b>11.60</b>
IBA @ 5000	12.51	13.22	11.21	15.90	14.58	<b>13.48</b>	9.75	11.25	9.00	11.75	12.50	<b>10.85</b>
NAA @ 1000	10.54	11.56	10.00	12.11	11.98	<b>11.23</b>	8.25	8.50	7.75	9.00	9.50	<b>8.50</b>
NAA @ 2000	11.22	12.10	10.03	13.30	12.56	<b>11.84</b>	8.50	8.75	8.00	9.25	10.00	<b>8.90</b>
NAA @ 3000	15.23	15.01	14.32	16.82	16.07	<b>15.49</b>	9.75	10.25	9.25	11.00	12.00	<b>10.45</b>
NAA @ 4000	20.06	21.33	19.38	24.06	22.71	<b>21.50</b>	12.25	12.00	10.00	13.50	14.50	<b>12.45</b>
NAA @ 5000	14.78	15.16	13.33	15.91	15.22	<b>14.88</b>	12.00	11.50	9.00	12.50	13.00	<b>11.60</b>
Control	11.22	11.86	10.60	13.35	12.94	<b>11.99</b>	8.00	8.75	7.75	9.25	10.00	<b>8.75</b>
<b>Mean</b>	<b>14.16</b>	<b>14.88</b>	<b>13.14</b>	<b>16.98</b>	<b>16.08</b>	<b>15.05</b>	<b>10.13</b>	<b>10.80</b>	<b>9.18</b>	<b>11.63</b>	<b>12.45</b>	<b>10.84</b>
	T = 0.089			T = 0.176*			T = 0.070			T = 0.138*		
	SE(d) M = 0.060			CD M = 0.118*			SE(d) M = 0.047			CD M = 0.093*		
	T×M = 0.199			T×M = 0.393*			T×M = 0.157			T×M = 0.310*		

\*Significant @ 5% level, Each value is the mean of four replications M1- Soil Sand: FYM (1:1:1) M2- Soil: Sand: FYM (1:1:1) M3- Soil: Coir pith (1:1) M4- Soil: Sand: Vermicompost (1:1:1) M5- Soil: Coir pith: FYM (1:1:1)

**Fig 3. Interaction of rooting hormone and rooting medium on survival per cent of *M. indica***



**Table 2. Effect of rooting hormone and rooting medium on Shoot length and number of roots of *M. indica***

Treatments	Shoot Length						Number of roots					
	M1	M2	M3	M4	M5	Mean	M1	M2	M3	M4	M5	Mean
IBA @ 1000	11.63	12.72	10.70	14.05	13.11	12.44	8.25	9.00	8.00	10.00	11.50	9.35
IBA @ 2000	15.03	16.00	14.23	18.37	16.22	15.97	11.00	12.25	10.50	13.50	14.25	12.30
IBA @ 3000	19.77	20.08	18.60	26.11	25.70	22.05	13.25	14.50	12.50	15.25	16.50	14.40
IBA @ 4000	13.82	14.73	12.20	16.81	15.81	14.67	10.50	12.00	9.25	13.00	13.25	11.60
IBA @ 5000	12.51	13.22	11.21	15.90	14.58	13.48	9.75	11.25	9.00	11.75	12.50	10.85
NAA @ 1000	10.54	11.56	10.00	12.11	11.98	11.23	8.25	8.50	7.75	9.00	9.50	8.50
NAA @ 2000	11.22	12.10	10.03	13.30	12.56	11.84	8.50	8.75	8.00	9.25	10.00	8.90
NAA @ 3000	15.23	15.01	14.32	16.82	16.07	15.49	9.75	10.25	9.25	11.00	12.00	10.45
NAA @ 4000	20.06	21.33	19.38	24.06	22.71	21.50	12.25	12.00	10.00	13.50	14.50	12.45
NAA @ 5000	14.78	15.16	13.33	15.91	15.22	14.88	12.00	11.50	9.00	12.50	13.00	11.60
Control	11.22	11.86	10.60	13.35	12.94	11.99	8.00	8.75	7.75	9.25	10.00	8.75
<b>Mean</b>	<b>14.16</b>	<b>14.88</b>	<b>13.14</b>	<b>16.98</b>	<b>16.08</b>	<b>15.05</b>	<b>10.13</b>	<b>10.80</b>	<b>9.18</b>	<b>11.63</b>	<b>12.45</b>	<b>10.84</b>
	T = 0.089					T = 0.176*	T = 0.070					T = 0.138*
	SE(d) M = 0.060					CD M = 0.118*	SE(d) M = 0.047					CD M = 0.093*
	T×M = 0.199					T×M = 0.393*	T×M = 0.157					T×M = 0.310*

\*Significant @ 5% level, Each value is the mean of four replications, M1- Soil Sand: FYM M2-Soil: Coir pith M3-Soil: Sand: Vermicompost M4- Soil: Sand: Vermicompost M5- Soil: FYM: Coir pith

**Table 3. Interaction of rooting hormone and rooting medium on Root length and number of leaves of *M. indica***

Treatments	Root length						Number of leaves					
	M1	M2	M3	M4	M5	Mean	M1	M2	M3	M4	M5	Mean
IBA @ 1000	12.76	13.04	11.70	15.93	14.13	<b>13.51</b>	5.50	6.50	5.00	8.25	7.75	<b>6.60</b>
IBA @ 2000	14.03	14.79	13.40	17.75	15.27	<b>15.05</b>	12.00	12.75	10.50	14.75	13.50	<b>12.70</b>
IBA @ 3000	20.37	22.91	18.76	26.03	23.03	<b>22.22</b>	15.75	18.50	14.75	22.50	20.00	<b>18.30</b>
IBA @ 4000	13.33	14.27	12.11	16.71	15.13	<b>14.31</b>	11.50	12.00	10.00	13.25	12.75	<b>11.90</b>
IBA @ 5000	11.70	12.89	10.99	12.33	12.76	<b>12.13</b>	5.25	6.00	5.00	7.50	6.75	<b>6.10</b>
NAA@ 1000	8.10	10.22	7.91	11.74	11.02	<b>9.79</b>	7.00	7.50	6.50	9.00	8.25	<b>7.65</b>
NAA@ 2000	8.22	10.78	8.04	12.83	11.94	<b>10.36</b>	7.50	8.00	7.00	9.25	8.50	<b>8.05</b>
NAA@ 3000	12.67	13.33	10.76	16.43	14.60	<b>13.55</b>	9.00	9.50	8.50	13.50	10.25	<b>10.15</b>
NAA@ 4000	19.38	20.07	18.20	23.01	22.43	<b>20.62</b>	11.50	12.00	10.75	17.50	14.75	<b>13.30</b>
NAA@ 5000	11.13	13.02	10.11	15.17	14.33	<b>12.75</b>	8.50	9.00	8.00	12.00	10.00	<b>9.50</b>
Control	8.55	9.65	7.24	12.78	11.33	<b>9.91</b>	6.50	7.00	6.00	8.50	8.00	<b>7.20</b>
<b>Mean</b>	<b>12.74</b>	<b>14.08</b>	<b>11.74</b>	<b>16.42</b>	<b>15.08</b>	<b>14.02</b>	<b>9.09</b>	<b>9.88</b>	<b>8.36</b>	<b>14.18</b>	<b>12.77</b>	<b>10.05</b>
	T = 0.085					T = 0.168*	T = 0.062					T = 0.122*
	SE(d) M = 0.057		CD M = 0.113*				SE(d) M = 0.041		CD M = 0.082*			
	TxM = 0.190					TxM = 0.375*	TxM = 0.138					TxM = 0.273*

#### 4. CONCLUSION

In the present study ideal size of 7 to 8 cm mini apical cuttings of mulberry (variety-V1) were subjected to hormonal treatment for root induction. Two growth regulators *viz.*, NAA and IBA at different concentrations (1000, 2000, 3000, 4000 and 5000 ppm) were used. Hormone treated mini cuttings were subjected to five different rooting medium *viz.*, Soil, Soil: sand: FYM (1:1:1), Soil: coir pith (1:1), Soil: sand: vermicompost (1:1:1) and Soil: coir pith: FYM (1:1:1). Out of two growth regulators, cuttings treated with IBA 3000 ppm registered superiority in overall performance *viz.*, survival percentage (68.80 %), sprouting percentage (82.39 %), rooting percentage (63.27 %), shoot length (22.05 cm), root length (22.22 cm), number of roots (14.40) and number of leaves (18.30) followed by NAA at 4000 ppm. In a holistic perspective, the study recommends that propagation of *M. indica* mini cuttings treated with IBA at 3000 ppm for better growth performance.

#### References

1. Parvez S, Ud-Din SN, Shafi S, Maqbool S, Bhat IA. To study the performance of summer raised rooted saplings in field at the end of different seasons, 2023.

2. Choudhary R, Malik SK, Chaudhury R, Rao AA. Optimized Recovery of Cryostored Dormant Buds of Mulberry Germplasm. *Plants*, 2023; 12(2), 225.
3. Tikader A, Vijayan K. Assessment of biodiversity and strategies for conservation of genetic resources in mulberry (*Morus* spp.). *Tree and Forest Biodiversity, Bioremediation, Biodiversity and Bioavailability*, 2010; 4(1), 15-27.
4. Samami R, Seidavi A, Eila N, Moarefi M, Ziaja DJ, Lis JA, Cappai MG. Production performance and economic traits of silkworms (*Bombyx mori* L., 1758) fed with mulberry tree leaves (*Morus alba*, var. Ichinose) significantly differ according to hybrid lines. *Livestock Science*, 2019;226, 133-137.
5. Saraçoğlu O, Erdem SÖ, Çekiç Ç, Yıldız K. Application of new vegetative propagation methods for black mulberry. *Uludağ Üniversitesi Ziraat Fakültesi Dergisi*, 2016; 30, 624-627.
6. Vijayan K, Tikader A, da Silva JAT. Application of tissue culture techniques for propagation and crop improvement in mulberry (*Morus* spp.). *Tree For Sci Biotechnol*, 2011; 5(1), 1-13.
7. Hartmann HT, Kester DE, Davies FT and RL Geneve,. *Plant Propagation: Principles and Practices*. 7th ed. Prentice-Hall. Englewood Cliffs, New Jersey: 2002; 363-365.
8. Parthiban KT, Seenivasan R. *Forestry technologies-a complete value chain approach*. Scientific Publishers. 2002.
9. Parthiban KT, Subbulakshmi V, Paramathma M, Devanand PS. Dardizing cutting size for clonal multiplication of *Jatropha curcas*. *Agricultural Science Digest-A Research Journal*, 2014; 34(2), 147-150.
10. Husen A, Iqbal M, Siddiqui SN, Sohrab SS, Masresha G. Effect of indole-3-butyric acid on clonal propagation of mulberry (*Morus alba* L.) stem cuttings: rooting and associated biochemical changes. *Proceedings of the National Academy of Sciences, India Section B: Biological Sciences*, 2014; 87, 161-166.
11. Ahalya BN. Genotype Environmental Interaction for Growth and Yield Parameters of Tree Mulberry Genotypes in Different Seasons and Bioassay Studies of Silkworm *Bombyx mori* L. *Mysore Journal of Agricultural Sciences*, 2014; 56(1).
12. Sajeewan RS, Singh SJ, Nataraja KN, Shivanna MB. An efficient in vitro protocol for multiple shoot induction in mulberry, *Morus alba* L variety V1. *Int. Res. J. Plant Sci*, 2011; 2(8), 254-261.
13. Mogili T, Rajashekar K, Balakrishna R, Kamble CK. Development of protocol for raising mulberry saplings from soft wood and semi-soft wood cuttings. *Advances in Plant Sciences*, 2011: 24(2), 443-446.
14. Kiruthika C, Susikaran S, Parthiban KT, Krishnamoorthy S V. Role of Auxins on growth of apical shoot cuttings of mulberry (*Morus indica* L.) using Mini clonal technology. *IJCS*, 2011;8(4), 1896-1899.
15. Wiesman Z, Riov J, Epstein E. Comparison of movement and metabolism of indole-3-acetic acid and indole-3-butyric acid in mung bean cuttings. *Physiologia Plantarum*, 1988; 74(3), 556-560.
16. Sun J, Li H, Chen H, Wang T, Quan JE, Bi H. The Effect of Hormone Types, Concentrations, and Treatment Times on the Rooting Traits of *Morus* 'Yueshenda 10' Softwood Cuttings. *Life*, 2023; 13(4), 1032.
17. Heloir MC, Kevers C, Hausman, J. F., & Gaspar, T. Changes in the concentrations of auxins and polyamines during rooting of in-vitro-propagated walnut shoots. *Tree Physiology*, 2023; 16(5), 515-519.
18. Husen A, Pal M. Effect of serial bud grafting and etiolation on rejuvenation and rooting cuttings of mature trees of *Tectona grandis* Linn. f. *Silvae Genetica*, 2003; 52(2), 84-87.
19. Rahdari P, Mohanna M, Asadi A. Effect of zinc sulfate and IBA and NAA on rooting of semihardwood cuttings of *Aralia* spp. and its' bio-environment effect. *Journal of Sciences and Techniques in Natural Resources*. 2010; 5 (1). 95-103.

20. Koyuncu F and Senel E. Rooting of Black Mulberry (*Morus nigra* L.) hardwood cuttings. J of fruit and ornament plant Res., 2003; 11: 53-57.
21. Rani S, Sharma A, Wali VK, Bakshi P, Ahmed S. The standardization of method and time of propagation in guava (*Psidium guajava*). Indian Journal of Agricultural Sciences. 2015; 85(9):38-45
22. Reddy KV, Reddy CP, Goud PV. Effect of Auxins on the rooting of fig (*Ficus carica* L.) hardwood and semi hardwood cuttings. Indian Journal of Agricultural Research, 2008; 42(1), 75-78.
23. Abdullah ATM, Hossain MA, Bhuiyan MK. Clonal propagation of guava (*Psidium guajava* Linn.) by stem cutting from mature stockplants. Journal of Forestry Research, 2006;17, 301-304.
24. Sukhjit K. Effect of different treatments of Indole3-butyric acid (IBA) on the rooting and growth performance of hardwood cuttings of Peach (*Prunus persica* L.).Agric. Sci. Digest, 2015; 35(1): 41-45.
25. Singh KK, Choudhary T, Kumar A. Effect of various concentrations of IBA and NAA on the rooting of stem cuttings of mulberry (*Morus Alba* L.) under mist house condition in Garhwal Hill region. *Indian Journal of Hill Farming*, 2014; 27(1), 125-131.
26. Sinha PS, Gangwar SK, Singh BD, Jaiswal J, Griyaghey UP. Evaluation of some elite Mulberry (*Morus alba* L.) varieties and NPK levels under partially irrigated conditions from sericulture view point. Indian Journal of Agricultural Research, 2001; 35(2), 71-78.
27. Husen A. Rejuvenation and adventitious rooting in coppice-shoot cuttings of *Tectona grandis* as affected by stock-plant etiolation. American Journal of Plant Sciences, 2011; 2(3), 370-374.
28. Husen A. Clonal multiplication of teak (*Tectona grandis*) by using moderately hard stem cuttings: effect of genotypes (FG1 and FG11 Clones) and IBA treatment. *Advances in Forestry Letters*, 2013; 2(2), 14-19.
29. Kalyoncu IH, Ersoy N, Yilmaz M, Aydın M . Effects of humidity level and IBA dose application on the softwood top cuttings of white mulberry (*Morus alba* L.) and black mulberry (*Morus nigra* L.) types. *African Journal of Biotechnology*, 2009; 8(16).
30. Habibi P, de Sa MFG, da Silva ALL, Makhzoum A, da Luz Costa J, Borghetti IA, Soccol CR . Efficient genetic transformation and regeneration system from hairy root of *Origanum vulgare*. *Physiology and molecular biology of plants*, 2016; 22, 271-277.
31. Leakey RRB, Chapman VR, Longman KA. Physiological studies for tropical tree improvement and conservation. Factors affecting root initiation in cuttings of *Triplochiton scleroxylon* K. Schum. *Forest Ecology and management*, 1982; 4(1), 53-66.
32. Singh KK, Rawat JMS, Tomar YK. Influence of IBA on rooting potential of torch glory *Bougainvillea glabra* during winter season. *Journal of Horticultural Science & Ornamental Plants*, 2011; 3(2), 162-165.
33. Singh KK, Krishan C, Singh KKK. Propagation of citrus species through cutting: A review. *Journal of Medicinal Plants Studies*, 2018; 6(1), 167-172.
34. Packialakshmi M, Jude Sudhagar R. Standardisation of rooting hormone in mini clonal technology of *Tectona grandis*. *International Journal of Chemical Studies*, 2019; 7 (3): 4398-4401
35. Ullah T, Wazir FU, Ahmad M, Analoui F, Khan MU. A breakthrough in guava (*Psidium guajava* L.) propagation from cutting. *Asian J. Plant Sci.*, 2005; 4, 238–243
36. Pallavi D, Sharma GL, Naik KL. Effect of IBA and NAA on rooting and growth of mulberry cuttings. *Int J Curr Microbiol App Sci*. 2018; 7(11):305-308
37. Singh KK. Effect of auxins and rooting media on rooting in stem cutting of mulberry (*Morus nigra* L.). *The Pharma Innovation Journal*, 2018; 7(11), 12-15.

38. Ghatnatti, SA. Studies on propagation of *Durandha plumeri* Jacq. Var Goldiana by stem cuttings with growth regulators under mist. M.Sc.(Agri.) Thesis, Univ. Agric. Sci., 1997.
39. Baroudi H, Makhoul G, Mahfoud H. The effect of IBA (Indole-3-Butyric Acid) dose applications on rooting of hardwood and softwood top cuttings of white mulberry (*Morus alba* L.) and black mulberry (*Morus nigra* L.). Syrian Journal of Agricultural Research. 2017; 5(2):2334.
40. Galavi M, Ali Karimian M, Mousavi SR. Effects of Different Auxin (IBA) Concentrations and Planting-Beds on Rooting Grape Cuttings (*Vitis vinifera*). Annual Review & Research in Biology. 2013; 3(4):517-523.
41. Kumar P. Genetic evaluation, Growth characterization and clonal propagation studies in *Melia dubia* Cav. Ph.D. Thesis, Tamil Nadu Agricultural University, Coimbatore,2011.
42. Wahab F, Nabi G, Ali N, Shah M. Rooting response of semi hard wood cuttings of guava (*Psidium guajava* L.) to various concentrations of different auxins. Online Journal of Biological Sciences. 2001; 1(4):184-87.
43. Siddiqui MI, Hussain SA. Effect of indole butyric acid and types of cuttings on root initiation of *Ficus hawaii*. Sarhad Journal of Agriculture, 2007; 23(4), 919.
44. Alam R, Rahman KU, Ilyas M, Ibrahim M, Rauf MA. Effect of Indole Butyric acid concentrations on the rotting of Kiwi cuttings. Sarhad Journal of Agriculture, 2007; 23(2), 293.
45. Husen A. Clonal propagation of *Dalbergia sissoo* Roxb. by softwood nodal cuttings: effects of genotypes, application of IBA and position of cuttings on shoots. Silvae Genetica, 2004; 53(1-6), 50-55.
46. Guleria V, Vashisht A. Rejuvenation and adventitious rooting in shoot cuttings of *Tectona grandis* under protected conditions in new locality of Western Himalayas. Universal Journal of Plant Science, 2014; 2(6), 103-1006.