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A study to evaluate the effect of using different concentrations of rooting hormone on root and shoot characters of apical cuttings of V1 mulberry (*Morus indica* L.) raised using mini clonal technology at nursery level

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Abstract

Research was done to standardize the rooting media and rooting hormone concentration in variety V1 apical shoot cuttings. Cuttings sown in sand: coir pith: FYM @1:1:1 ratio rooted 5cm deep and 15cm in length, giving the cuttings a higher chance of survival. Between treated and untreated cuttings, there were notable differences in shoot, root characters and the quantity of main roots. Cuttings treated with 3000 ppm IBA showed enhanced root number (no's), shoot length (cm), no of leaves (no's) and root length (cm). In apical shoot cuttings with active node, the number of roots and success rate of rooting were found to be maximum. The best outcomes for the mini-cuttings' root and shoot growth came from combining appropriate rooting medium with cuttings of the right kind and concentration of IBA, as per the improved methodologies. The findings suggested that V1 apical cuttings with healthy terminal bud might be utilized effectively in mini clonal technology to create saplings with potential vigour.

Keywords: Auxins, apical cuttings, biochemical, mulberry, mini clonal technology, V1 variety

Introduction

A perennial plant in nature, mulberry (*Morus alba* L.) is raised for its foliage, which is used as a seasonal crop to feed silkworms (*Bombyx mori* L.) (Datta, 2000) [3].

Being perennial in nature, mulberry plantations continuously produce high-quality leaves for around 15 to 20 years after they are established (Hawramee *et al.*, 2019) [10]. Popularly, mulberry plants are propagated vegetatively. Thus, an effective propagation technique is essential to prosper the future of sericulture as well as saving farmers' money. For this experiment, the high-yielding mulberry cultivar Victory1 (V1) was chosen while taking all of these factors into account. Because of its poor viability and high heterozygosity (Hartmann *et al.*, 1990) [6], seedling propagation was not popular for the routine multiplication of mulberries; as a result, seedling production was limited to breeding trials (Vijayan *et al.*, 1997) [37].

Stem cuttings are the main method used to propagate mulberries. Cuttings from stems are utilized as propagation material, based on the type of semi-hardwood. In addition to the edaphic and climatic circumstances, other elements that affect roots and the subsequent survival of cuttings include the type of wood, age of the stock plant, active lateral buds, and planting time. Hard wood cuttings from branches that are 6 to 8 months old after pruning are often the most widely used and conventionally advocated for planting (Dandin *et al.*, 2003) [2]. The farming community has benefited in recent years from the introduction of improved mulberry cultivars, which have resulted in maximum leaf output. But because of seed cuttings and sapling materials for transplantation are not readily available, farmers are finding it difficult to use these advanced mulberry types and make them promptly available (Mogili *et al.*, 2011) [18].

Thus, the most essential need is to raise nurseries for any type of farming crop, whether it be agricultural, horticultural, or sericulture. In order to successfully develop a standing crop, nurseries were raised with great care in limited space by giving all essential inputs, such as manures, fertilizers, and timely watering in a protective manner for rapid, healthy, and with established shoot and root system. The presence of competing weeds, soil temperature, and soil moisture all have a significant impact on the vigour of the sapling and its ability to sprout.

Using soft or green wood cuttings from the seed wood gardens, Sabarish (2017) [29] tried to produce mulberry saplings in 60 to 70 days. After being administered with root stimulating hormone (IBA) and planted in soil wrapped in bamboo-framed polythene sheets, the cuttings took 45 days to produce roots. They have succeeded in achieving 50 to 75% survival using this procedure, but they have also suffered the same challenges as traditional cutting plants (Bharathi *et al.*, 2022) [14].

It is now essential to keep in mind the challenges associated with raising traditional (bed) saplings when executing "Clonal production of saplings with root trainer methods" in order to produce established clonal saplings on a large scale in less time (within 100 days), with less laborious work, at a lower cost, and that the farming community can easily transfer and transplant (Prakash *et al.*, 2017) [25]. A clone is made up of vegetatively propagated saplings that were created from a single superior plant that has all the desired traits (Ruppert, 1974) [26]. As a result, every clone's clonal saplings are identical, true to type, and possess every genetic trait of the mother tree (Parthiban *et al.*, 1999) [22]. The genetic characteristics of each clone vary from one another, and each clone reflects a certain genotype (Parthiban and Seenivasan, 2017) [14]. Thus, the study was started with the goal to introduce the same technology to propagate mulberry.

Materials and Methods

At the Forest College and Research Institute in Mettupalayam, which is situated 300 meters above mean sea level at latitude 11°9'N and longitude 77°56'E, where the experiment was undertaken. Apical shoot cuttings from variety V1 were utilized as the plant material chosen for the study. Mini cuttings were raised in a mist chamber and a low-cost poly tunnel. During the early morning, sterile pruning scissors were used to remove the mini-cuttings from the mother plants. Following harvesting, they were screened using a calibrated vernier calliper to determine the appropriate diameter (1-1.5 cm) and length (15 cm) using a scale. The mini-cuttings that were chosen were subsequently stored in an ice box to avoid desiccation damage and to facilitate their transit to the laboratory. They were subjected to a 0.1% systemic fungicide treatment for 15 minutes. Later, the cuttings were treated at concentrations of 1000, 2000, 3000, 4000 and 5000 parts per million (ppm) with several rooting hormones, namely IBA and NAA. The weighed quantity of each hormone was first dissolved in sodium hydroxide solution, then the volume was adjusted with distilled water. The stock solutions of these hormones were also tested in powder base application. The rooting medium made of coir pith: sand: FYM (1:1:1) was planted with the treated cuttings and found to perform better than other treatments. The cuttings were then inserted into the mist chamber and poly tunnel. The cuttings were periodically sprayed in the mist chamber (30 minutes at a time) to maintain a temperature of 25 to 30 °C or 15 to 20 °C during the day and night. By employing rose cans to spray water intermittently, the average temperature and relative humidity of the poly tunnel were kept at 25 to 35 °C and 70 to 80 percent

respectively.

Following a period of sixty days, the cuttings were delicately harvested from the rooting medium and measurements were made of the root numbers, root length, shoot length, and no of leaves. The rooted cuttings were then placed in polythene bags (15 x 25 cm) that were filled with soil and Farm Yard Manure (5:1), and they were kept in a shade house for a period of fifteen to twenty days for hardening. In each treatment, biometric data related to the following attributes were recorded *viz.*, number of roots, root length, number of roots, and no of leaves.

The Design of Statistics

A completely randomized design (CRD) with three replications was used to set up the experiment. The significance of the data was assessed using Analysis of Variance (ANOVA) in accordance with the protocol developed by Panse and Sukhatme (1978) [20].

Results and Discussion

In this study, rooting hormones (IBA and NAA) at different concentrations 1000, 2000, 3000, 4000 and 5000 ppm were found to have a substantial impact.

Effect of rooting hormone on shoot length of V1 mulberry apical cuttings

When comparing all the treatments with varying IBA and NAA dosages, IBA at 3000 ppm registered the highest shoot length. At 3000 ppm, the shoot length was 23.75 cm with IBA application followed by 19.80 cm @ 4000 ppm. With NAA dosages, 18.75cm @ 4000 ppm were found to be maximum among all dosages in V1 variety. The administration of IBA and NAA triggers root initiation and shoot development. According to Kalyoncu *et al.* (2009) [15], IBA application in black mulberry had the enhanced shoot length. Cuttings of black mulberry treated with high IBA dosage in bunch planting leads to increased shoot length, according to Koyuncu and Senel's (2003) [3].

An increase in IBA concentrations triggered the shoot length of oleander plants to maximize as observed by Habibi (2010) in his experiments. High concentration IBA application caused the shoot length decrease considerably. In specific *Triphlochiton* sp. clones, increased dosage of rooting hormone (200 µg per cutting) in the study curtailed the root and shoots in cuttings (Leakey *et al.*, 1982b) [17]. The experiment clearly depicts, shoot length found minimum @ 3000 ppm concentration. Similar results has been observed by Husen *et al.* (2015) [9], Singh *et al.* (2011) [33] in *Bougainvillea glabra*, Singh *et al.* (2013) [32] in Citrus lemon cv. Cuttings, Singh *et al.* (2014) [31] and Packialakshmi and Sudhagar (2019) [23] in teak apical cuttings, clearly shows that the application of rooting hormone could substantially increased the shoot length in many related species.

Effect of rooting hormone on number of roots of V1 mulberry apical cuttings

Highest number of roots were registered in IBA compared to NAA when both hormones were used in the experiment. After 90 DAP, IBA at 3000 ppm registered the more number of roots 16.75 no's followed by 15.33 no's @ 4000 ppm. For NAA, at 4000 ppm (15.20 no's) high number of roots were noticed followed by 12.20 no's @ 5000 ppm. In *Psidium guajava*, mini cuttings treated with IBA application at 3000 ppm and above concentration recorded more roots, which may be due to the nominal hormonal impact that accumulates essential substances and triggers their downward movement (Rani *et al.*, 2018) [27]. According to (Pallavi *et al.*, 2018) [19], fluctuation in dosage

levels with relation to the number of roots will led to the varietal and climatic changes in the particular location. Numerous species showed maximum rooting in cuttings as a result of auxin treatment triggers cambium activity which are in line with Ullah *et al.* (2005) [35].

Effect of rooting hormone on root length of apical cuttings of V1 mulberry variety

The current experiment clearly states that IBA registered the maximum root length among all treatments. The highest root length recorded at 90 DAP was 27.25 cm in IBA application at 3000 ppm followed by 23.50 cm in IBA at 4000 ppm similarly 19.80 cm in NAA at 4000 ppm followed by 17.25 cm in NAA at

5000 ppm concentration. Similar enhanced hormonal effect was noticed by Kumar (2018) [12] in mulberry, leading to enhanced root length. Highest root length was registered by Ghatnatti in 1997 [5]. This was attributed to increased hormone activity, which led to hydrolysis and transport of carbohydrates towards the cuttings base, which resulted in cell division and elongation at particular site. According to Baroudi *et al.* (2017) [1], mulberry (*Morus alba*) softwood cuttings treated with 2000 ppm and high concentrations of IBA showed good root length, root number, and rooting percent. The results of Kumar (2011) [16] in *Melia dubia* and Galavi *et al.*, 2013 [4] in *Vitis vinifera* well supported the research findings.

Table 1: Effect of IBA on growth attributes of *Morus indica*

Treatments (IBA)	On 90DAP			
	Shoot length (cm)*	No of roots/plant (no's) *	Root length (cm) *	No of leaves/plant (no's) *
IBA@1000 ppm	12.25 ^e (3.64)	9.80 ^e (3.28)	15.75 ^e (4.09)	15.10 ^e (4.01)
IBA@2000 ppm	13.00 ^d (3.74)	11.50 ^d (3.53)	17.80 ^d (4.33)	16.80 ^d (4.21)
IBA@3000 ppm	23.75 ^a (4.97)	16.75 ^a (4.21)	27.25 ^a (5.31)	23.75 ^a (4.97)
IBA@4000 ppm	19.80 ^b (4.56)	15.33 ^b (4.04)	23.50 ^b (4.94)	20.10 ^b (4.59)
IBA@5000 ppm	17.33 ^c (4.28)	12.75 ^c (3.70)	21.33 ^c (4.72)	18.33 ^c (4.39)
Control	0.00 (1.00)	0.00 (1.00)	0.00 (1.00)	0.00 (1.00)
SEd	0.08	0.05	0.09	0.07
CD (.05%)	0.19	0.13	0.21	0.15

Significant @ $P=0.05$ level, Each value is the mean of four replications

*Values are square root transformed values

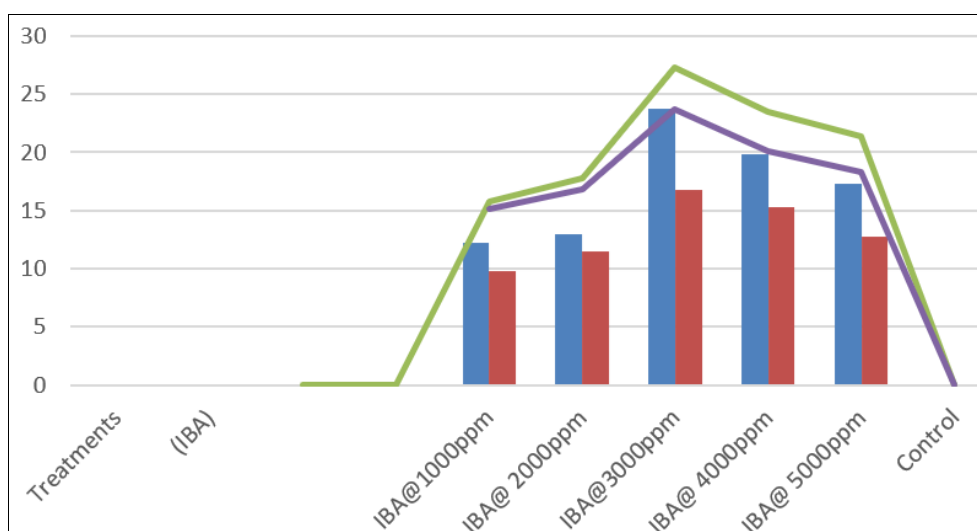


Fig 1: Effect of IBA on growth attributes of *Morus indica*

Table 2: Effect of NAA on growth attributes of *Morus indica*

Treatments (NAA)	On 90DAP			
	Shoot length (cm) *	No of roots/plant (no's) *	Root length (cm) *	No of leaves/plant (no's) *
NAA@1000 ppm	12.00 ^e (3.60)	9.10 ^e (3.17)	14.33 ^e (3.91)	9.20 ^e (3.19)
NAA@2000 ppm	14.33 ^d (3.91)	10.33 ^d (3.36)	17.20 ^d (4.26)	11.10 ^d (3.47)
NAA@3000 ppm	16.75 ^c (4.21)	11.50 ^c (3.53)	19.80 ^c (4.55)	15.33 ^c (4.04)
NAA@4000 ppm	18.75 ^a (4.44)	15.20 ^a (4.02)	23.25 ^a (4.92)	19.80 ^a (4.55)
NAA@5000 ppm	17.80 ^b (4.33)	12.20 ^b (3.63)	22.50 ^b (4.27)	17.25 ^b (4.27)
Control	0.00 (1.00)	0.00 (1.00)	0.00 (1.00)	0.00 (1.00)
SEd	0.07	0.06	0.08	0.08
CD (.05%)	0.16	0.13	0.17	0.19

Significant @ $P=0.05$ level, each value is the mean of four replications

*Values are square root transformed values

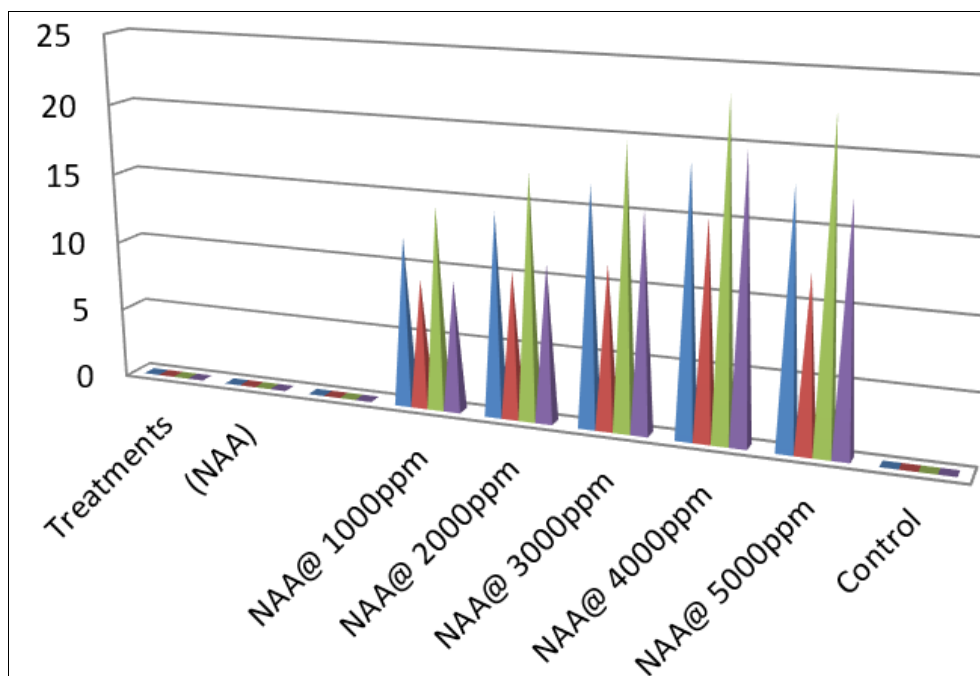


Fig 2: Effect of NAA on growth attributes of *Morus indica*

Effect of rooting hormone on number of leaves of apical cuttings of V1 mulberry variety

From the experiment, IBA at 3000 ppm recorded highest number of leaves of 23.75 no's at 90 DAP among IBA and NAA dosages followed by IBA at 4000 ppm registered more number of leaves of 20.10 no's. NAA at 4000 ppm recorded more number of leaves of 19.80 no's followed by 17.25 no's @ 5000 ppm. According to Pallavi *et al.* (2018) [19], there may be increased amount of roots and branches in mulberry cuttings at maximum ppm concentration, which led to more number of leaves. The influence of IBA on the more number of leaves arises from the activation of shoot and root growth, which results in an enhanced more nodes and the subsequent formation of additional leaves in *Psidium guajava* L., Kiwi cuttings (Riaz *et al.*, 2007) [28], *Ficus Hawaii* (Ismail and Asghar, 2007) [11] which strengthens the current findings (Wahab *et al.*, 2001) [37].

Conclusion

It is concluded that IBA@3000 ppm and NAA@4000 ppm reported the highest values for significant root features in mini clonal technology based on the findings of the current study. As of right now, the work on mini clonal propagation is quite less available for clonally propagating V1 mulberry variety from apical shoot cuttings. As a result, the current study has standardized the rooting hormone and its concentration, which is found to be a promising technique that could eventually replace traditional stem cutting (utilizing lateral buds to regenerate plants) propagation in V1 variety by producing large quantities of high-quality planting material quickly and with a high success rate.

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