__Review Article__

Effect of IBA on Performance of Mulberry Saplings: A Review

ABSTRACT

Mulberry (*Morus sp.*), which is the only food to silkworm is of great importance to sericulture industry. It can be propagated by seeds, cuttings, budding and grafting. Propagation by cuttings has distinct advantages over the other methods due to high survival rate and quick establishment. The use of growth regulators like IBA for cuttings helps to induce root formation, increase the root number and helps in mobilization of carbohydrates for root growth. The saplings treated with IBA have higher survival rate, number of leaves, length of shoot and rooting percentage.

Keywords: cuttings, auxin, IBA, mulberry.

1. INTRODUCTION

Sericulture has been practiced in India from very ancient time. It is labour oriented commercial activity, which provides high employment and good returns. According to International Sericultural Commission (ISC), India ranks second in silk production and the production of silk in 2021 is 34,903 MT. As a dynamic small-scale industry the employment potentiality of the sericulture industry is extensive. It helps to provide not only periodic return within a short period of time, but also guarantees potential employment opportunities to the sericulture family around the year. It is one of the stable enterprises which provides a continuous flow of returns in the southern states of India like Karnataka, Andhra Pradesh, Tamil Nadu, Kerala and Maharashtra throughout the year. Mulberry nursery management has gained the position of a commercial venture with its short gestation period and low investment. Few entrepreneurs have made it as a year-round activity with established supply chain across the Southern states.

Propagation is a common practice employed in all plants in order to obtain healthy and resistant plants. Different modes of propagation of mulberry are important to cultivate in large scale. Usually seeds, cutting, budding and grafting propagation methods are used in mulberry (*Morus* sp.). Rooting of the cutting is one of the possible techniques for vegetative propagation and it is observed that cuttings play a crucial role in the rooting of important species of some fruit species and clonal rootstocks. Propagation of mulberry from semi hard wood cuttings has distinct advantages over the direct plantation of cuttings, majorly higher survival rate due to already developed root system. Saplings enable quick establishment and vigorous growth. Realizing the importance of the initial establishment of garden by using saplings, farmers are preferring saplings for the establishment of the mulberry garden.

Endogenous and exogenous factors such as genotype, timing, cutting types, rooting environment and the age of the parent material from which cuttings were obtained influence the growth of the saplings. Application of exogenous plant growth promoting substances recommended for promoting adventitious roots in stem-cutting propagation for overcoming these problems is an alternative [1]. Generally, auxin and auxin like compounds are used to induce root formation, increase the root number and root length. Auxins are synthesized by Indole-3pyruvic acid (IPA) pathway by using the precursor tryptophan. Plants largely produce auxin in shoot tips and translocate to roots. Auxins, mainly used to induce the apical dominance, fruit development and lateral root formation. Generally, auxin and auxin like compounds are used to induce root formation, increase the root number and root length [2]. Treatment of auxins to cuttings causes physiological changes during the adventitious root formation which helps in mobilization of carbohydrates from leaves and upper stem, and accelerates their transport to the rooting zone. Among auxin compounds, indole-3-butyric acid (IBA) is the most widely used root promoting chemical in the nursery trade, because it is non toxic over a wide range of concentrations.

2. REVIEW OF SOME KEY RESULTS OF EARLIER REPORTED RESEARCH WORKS

2.1. Studies on biochemical changes induced by IBA on mulberry cuttings

Rooting of cuttings is influenced by a number of external and internal factors and the rooting response also varies with variety, planting season and the nature and concentration of harmones present in cuttings [3]. Application of growth regulators on cuttings was superior to the grafting process and was most economical for large scale multiplication of mulberry plant [4]. Leopold [5] stated that auxins and their compounds are used for rooting of cuttings but the most commonly used is IBA. The success of this compound is due to its low auxin activity and its slow degradation by auxin destroying enzymes. Auxins regulate various aspects of plant growth and development by affecting numerous processes including cell division, cell enlargement and differentiation [6]. According to Mitra and Bose [7], application of auxin has been found to enhance the histological features like formation of callus and tissue and differentiation of vascular tissue in *Boerhavia diffusa* L. Skoog [8] found that auxin moves upward in the xylem with the transpiration stream, then moves laterally into the surrounding tissues, and then is re- exported by normal polar basipetal transport. The downward movement appeared to consist of cell-to-cell movement in non-vascular tissues (at a faster rate) and in the phloem. Nanda et al. [9] have reported that high rooting success of IBA treated cuttings might be attributed to the fact that auxin induces hydrolysis and mobilization of nutritional factors to the site of application thereby promoting root initiation in stem cuttings. Auxin results in the breakdown of starch into soluble sugars bulk of which was used in rooting process. Auxins help in mobilization of carbohydrates in leaves and upper stem, and accelerates their transport to the rooting zone. The oxidative enzymes, widely distributed in higher plants, have special significance during the rooting. Changes in the pattern of IAA oxidase and peroxidase (POX) activities have been proposed as being the biochemical markers for the successive rooting phases [10]. Husen [11] identified that application of auxin to cuttings of *Grewia optiva* causes metabolic changes during the adventitious root formation, which consists of three successive but independent phases, namely induction, initiation and expression. The induction phase comprises of molecular and biochemical events without visible changes, the initiation phase is characterized by cell division and root primordia organization and the expression phase denotes the intra-stem growth of root primordia and the emergence of roots. Since rooting is a high-energy-demanding process, rooting ability of cuttings has been frequently discussed in relation to soluble and storage carbohydrate contents. Torrey [12] found that the hormone present in rooting part stimulates root initiation and translocates these harmones via the xylem sap and probably on occasions via the phloem to other part.

According to Aminah et al. [13] application of auxin (IBA) significantly increased the rate of root emergence in single node leafy stem cuttings of *Shorea leprosula*. The IBA can break down and decompose quickly to a low concentration which is appropriate to change the root meristem to root. Ahkami et al. [14] have proposed that auxin may enhance auxin concentration in the rooting zone to much higher levels so that the balance between responses at sink-establishment level (modifying the carbohydrate influx) versus root-development level (modifying the carbohydrate utilization) differs. However, enhanced sugar level in the rooting zone of cuttings caused by auxin treatment may be attributed to increase in starch hydrolysis or increased sugar transport towards the rooting zone. Johnson and Bonner [15] described the uptake of uptake of auxins by plant tissue consisting of three separable processes. The first, completed in 20 to 30 minutes, resembles a diffusion into the tissue in the sense that auxins taken up by this process are readily leached to the outside solution. The second, also a rapid process, resembles a binding, since the bound auxin can be removed by exchange for unlabeled auxin. The third is a continuous absorption, maintained at a steady rate for several hours. They also determined auxins could be accumulated by the tissue to a concentration higher than the external concentration and the rate of continuous uptake was inhibited by low temperature and metabolic inhibitors. According to Das et al. [16] auxins promote starch hydrolysis and mobilize sugars and nutrients to the cutting base. During cell division and auxin transport, auxins act primarily through selective proteolysis and cell wall loosening with receptor protein transporting inhibitor response 1 and auxin-binding protein 1. Gyana [17] stated that adventitious roots of *Camellia sinensis* were obtained in three distinct phases i.e., induction (0–12 days), initiation (12–14 days) and expression (14–18 days). IAA-oxidase activity of IBA treated cuttings increased slightly as compared to control. The activity was found to decrease during induction and initiation phases and increase during expression phase. The peroxidase activity in IBA-treated cuttings increased up to initiation phase and declined at the expression phase. Polyphenoloxidase activity was found to increase both in IBA-treated and control cuttings during induction and initiation phase but declined slowly during expression phase. Total phenolic content was higher in IBA-treated cuttings, particularly in initiation and expression phases and it also correlated with peroxidase activity. Phenolics might be playing key role for induction of adventitious rooting and phenolic compounds can be used as rooting enhancers in tea. Thimann and Koepfli [18] summarized that, the polar transport of auxin occurs primarily in a basipetal direction, with the shoot apices serving as the primary source of endogenous auxin for the entire plant. Polar transport contributes to the creation of an auxin gradient from the shoot to the root, with the auxin gradient affecting various physiological processes.

2.2 Review based on effect of IBA on shoot outgrowth

Singh et al. [19] treated stem cuttings of *Morus alba* with 1000, 1500 and 2000 ppm of IBA and NAA solutions by quick dip method. Among all the treatments, number of sprouted cuttings, length of the roots/cutting, percentage of rooted cutting, length of longest sprout of roots were higher in cuttings treated with IBA (2000 mg/l). Boschini and Rodriguez [20] examined that the use of IBA (4000 ppm) caused an 11 to 15% increase in total sprouting of mulberry cuttings because of early completion of physiological process involved in rooting and sprouting of cuttings. Ismail and Asghar [21] conducted an experiment to study the effect of different concentrations of IBA on cuttings of *Ficus hawaii* that were treated with 1000, 2000, 3000, 4000, 5000 ppm and 0 ppm (control). The results showed that the treatment of IBA produced significant variation. Maximum sprouting (43.7%) leaves per plant (63), plant height (37.46cm), shoots per plant (13), leaf area (19.33 cm^2) and shoot thickness (0.57 cm), were recorded in cuttings treated with 4000 ppm IBA. Barde et al. [22] treated cuttings of pomegranate with IBA at 2000 ppm which resulted in maximum number of shoots per cutting (2.80), number of leaves per cutting (38), dry matter percentage of leaves (46.7 %) and number of roots per cutting (10.83). According to Pallavi et al. [23] higher level of survival percentage of mulberry saplings is due to better development of root and shoot system of mulberry saplings in IBA (2000 ppm) treated cuttings. Singh and Singh [24] stated that appropriate planting time, application of IBA as well as genetic makeup of genotype might have played some role in augmenting the number of leaves per sapling in mulberry. Neelima et al. [25] conducted an experiment consisting of four different concentrations of IBA (500, 1000, 1500 and 2000 ppm) each along with control (Distilled water treatment) were treated for root initiation in cuttings of jasmine. The results revealed that growth regulator IBA had significant effect on survival and rooting performance of jasmine. The maximum survival percentage (88.33%) of rooted cuttings, less days to sprouting per cuttings (8.25), maximum number of buds per cutting (2.75), number of leaves per rooted cutting (10.58) and length of shoot per rooted cutting (3.30 cm) were recorded maximum with treatment of IBA at 1500 ppm. While the maximum number of main roots per rooted cutting (9.33) and length of root per rooted cutting (5.10 cm) was also recorded in cuttings treated with 1500 ppm of IBA.

2.3 Review based on effect of IBA on root growth

Auxins and its components gave better results for rooting, among which IBA was highly effective for rooting of the cuttings and this exogenously applied IBA is slowly destroyed by auxin destroying enzyme system thereby available to plant for a longer period of time [26]. Chadwick and Kiplinger [27] demonstrated that depending upon species, auxin treatment could enhance rooting percentage and quality of the root system. Plants normally propagated by softwood cuttings rooted in less time compared to non treated cuttings. Chaithanya et al. [28] conducted an experiment to find out the effect of IBA (auxin) concentration and size of cuttings on rooting and per cent establishment of *Jasminum sambac* stem cuttings. Three node cuttings treated with IBA at 2000 ppm exhibited significant increase in rooting percentage, root length, root number, root fresh weight, maximum sprout diameter, fresh and dry weight of rooted cuttings. Whereas four node cuttings treated with 2000 ppm of IBA and three node cuttings treated with 3000 ppm of IBA have obtained more number of sprouts and maximum number of leaves per cutting. Among all the treatment combinations, three node cuttings treated with IBA @ 2000 ppm has shown best results. Singh et al. [29] reported that cuttings treated with 5000 ppm of IBA planted in August, performed the best in all aspects, rooting percentage, number of primary roots, secondary roots, length of longest root, fresh and dry weight of root of mulberry saplings. Fotader et al. [30] screened six promising mulberry varieties for rooting and induction of rooting in hardwood cuttings using three growth regulators, *viz.* indole butyric acid (IBA), indole acetic acid (IAA) and naphthalene acetic acid

(NAA), in two concentrations (50 and 100 ppm). Among them, Chinese white showed the highest rooting percentage due to its good rootability and short gestation period. It was also found to be the most suited variety for temperate conditions. IBA at 100 ppm proved to be an effective treatment for inducing rooting in mulberry varieties which otherwise were having pre-potency for rooting. The varieties which have less pre-potency showed less expression on rooting even with the use of any of the growth regulators. Singh and Singh [24] examined that IBA at 4000 ppm showed highest percentage of rooting, number of primary roots, length of longest primary root in bougainvillea cuttings. IBA translocates poorly and remains near the site of application, and so it was found to be one of the best rooting stimulator. Hartmann et al. [31] stated that IBA induces basipetal transport of assimilates, with sink strength successively enhanced by increased IBA concentration. This process may account for the increase in number of roots per rooted cutting with increasing IBA concentration.

The difference among various concentrations of IBA could be related to such factors as higher stability and slow rate of conjugation, so that higher IBA concentration is required to induce more rooting and this will be available over a longer period of time [32]. According to Husen and Pal [33] higher concentration of IBA (3000 ppm) resulting in maximum rooting percentage (87 %) and reported that in IBA treated cuttings, enzyme IAA-oxidase and peroxidase (POX) help in auxin catabolism and in triggering the root initiation processes, the former is basically having a role in triggering and initiating the root primordium, while the latter in both the initiation and elongation processes. Polat [34] evaluated indole butyric acid (IBA) effect on rooting of four local cultivars of mulberry *Morus* spp. cvs. Beyrudi, Hatuni, Sami and Yabani. Cuttings of 20-25 cm in length during the dormant period from one-year-old shoots. Basal sections (1-2 cm) of the cuttings were treated with various concentrations of IBA solution for 10 s before planting. After three months of planting, rooting percentage, root number per cutting, average root length (cm) were determined. Most favorable rooting results were obtained from cuttings treated with 5000 ppm IBA. Highest rooting, 31.7%, was obtained with 5000 mgL^{-1} IBA in 'Beyrudi'. Kalyoncu et al. [35] studied the effect of Indol-3-butyric acid (IBA) on softwood top cuttings of two black mulberry and one white mulberry types. The highest rooting percentage was determined in (black mulberry) with 2000 and 3000 ppm IBA doses application (100%). Chumpookam et al. [36] observed the effect of IBA and NAA on rooting and axillary shoot outgrowth of 'Chiangmai 60' mulberry (*Morus alba* L.) stem cutting. They found that the stem cutting which was treated with IBA and NAA did not show significant increase in rooting percentage but 1000 ppm of IBA was optimum for the growth of 'Chiangmai 60' mulberry propagated by stem cutting.

3. CONCLUSION

To overcome the shortage of propagation material of mulberry cuttings due to climate change and outbreak of pest and diseases, it is necessary to treat the mulberry cuttings with optimum concentration of indole-3-butyric acid (IBA). As treated saplings showed highest rooting percentage, survival rate, number of leaves and they also induce the biochemical changes in cuttings which is required for root formation. Hence, by using IBA on mulberry cuttings the production of saplings on a large scale can be more economical and taken up as one of the income generating activities.

References

1. Sulaiman MK. The effect of auxin IBA and kinetin in budding success percentage of mulberry (Morus sp.). Int J Pure Appl Sci Technol. 2012;13(1): 50-56.
2. Geetha T, Murugan N. Plant growth regulators in mulberry. Annu Res Rev Biol. 2017;13(3): 1-11.
3. Hartmann HT, Kester DE, Davies FT, Geneve RL. Plant propagation: principles and practices. Upper Saddle River, NJ: Prentice Hall. 2002;253-257.
4. Rao LSP, Khan AA. Vegetative propagation of Japanese mulberry varieties by use of growth regulators. Indian J Seric. 1963;1(3): 7-23.
5. Leopold AC. Auxin and plant growth. Uni. of California Press, Bakeley Los Angels. 1960.
6. Woodward AW, Bartel B. Auxin: regulation and interaction. Ann. Bot. 2005;95: 707-735.
7. Mitra GC, Bose N. 1954, Rooting and histological responses of detached leaves to Indole Butyric Acod with special reference to Boerhavia diffusa L. Phytomorphology. 1954;7: 370.
8. Skoog F. Absorption and translocation of auxin. J Bot. 1938;25: 361-372.
9. Nanda KK, Anand VK, Kumar P. Investigation on the use of auxins in vegetative reproduction of forest plants. Indian Forester.1970;96(3): 171-187.
10. Li Sw, Xue L, Feng H. IBA induced changes in antioxidant enzymes during adventitious rooting in mung bean seedlings. Envrion Exp Bot. 2009;66: 442-450.
11. Husen A. Changes of soluble sugars and enzymatic activities during adventitious rooting in cuttings of Grewia optiva as affected by age of donor plants and auxins treatments. Amer J Plant Physiol. 2012;7: 1-16.
12. Torrey JG. Root hormones and plant growth. Ann Rev Pl Physiol. 1976;27: 435-459.
13. Amina H, Dick JM, Leakey RRB, Grace J, Smith RI, Effect of indole butyric acid (IBA) on stem cuttings of Shorea leprosula. Forest ecology and management. 1995;72(2): 199-206.
14. Ahkami AH, Melzer M, Ghaffari MR, Pollmann S, Javid MG, Shahinnia F, Hajirezaei MR, Druege U. Distribution of indole-3-acetic acid in Petunia hybrida shoot tip cuttings and relationship between auxin transport, carbohydrate metabolism and adventitious root formation. Planta. 2013;238: 499–517.
15. Johnson MP, Bonner J. The uptake of auxin by plant tissue. Physiol Plant. 1956;9: 102-118.
16. Das P, Basak U, Das A. Metabolic changes during rooting in pre-girdled stem cuttings and air layers of Heritiera. Bot Bull Acad Sin. 1997;38: 91-95.

17. Gyana RR. Effect of auxins on adventitious root development from single node cuttings of Camellia sinensis (L.) kuntze and associated biochemical changes. Plant Growth Regulation. 2006;48: 111-1117.
18. Thimann KV, Koepfli JB. Identify of the growth promoting and root forming substances of plants. Nature. 1935;135: 101-102.
19. Singh KK, Choudhry 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 J. Hill Farming. 2014;27(1): 125-131.
20. Boschini C, Rodriguez AM. The stimulation of rooting in mulberry (Morus alba L.) with IBA. Agromania Mesoamericana. 2002;13(1): 19-24.
21. Ismail SM, Asghar HI. Effect of indole butyric acid and types of cuttings on root initiation of Ficus Hawaii. Sarhad Journal of Agriculture. 2007;23(4): 919-925.
22. Barde P, Tiwari R, Kanpure RN, Baghel BS, Kumawat BR. Effect of biofertilizer and growth regulators on rooting and growth of pomegranate cuttings. Annals Pl Soil Res. 2010;12(1): 46-47.
23. 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.
24. Singh AK, Singh VS. Influence of wood maturity and auxins on the regeneration of Bougainvillea cuttings. Prog Hort. 2002;34(2): 196-199.
25. Neelima N, Neeraj S, Gaurav S, Jitendra KS. Effect of different IBA concentration on survivability and rooting of jasmine (Jasminum sambac L.) stem cuttings. Journal of Pharmacognosy and Phytochemistry. 2018;614-617.
26. Stoutemeyer VT. Humidification and the rooting of green wood cutting of different plants. Proc Amer Soc Hort Sci. 1942;40: 301-304.
27. Chadwick LC, Kiplinger DC. The effect of synthetic growth substances on the rooting and subsequent growth of ornamental plants. Proc Amer Soc Hort Sci. 36: 809-816.
28. Chaitanya RP, Chandrasekhara RC, Suchitra, V, Salomi S. Effect of IBA concentration and size of cutting on rooting of Jasminum sambac. Green Farming. 2015;6(4): 888-892.
29. Singh KK, Dev KJ, Mehta SK. Rootability of hardwood cuttings of mulberry (Morus alba L.) influenced by planting time and growing conditions under valley condition of Garhwal Himalayas. Plant Archives. 2015;15(2): 1031-1036.
30. Fotadar, RK, Ahsan MM, Dhar KL, Dhar A. Screening of mulberry varieties for rooting and induction of rooting by the use of growth regulators. Sericologia. 1990;30(3): 347-367.
31. Hartmann HT, Kester DE, Davies FT, Geneve RL. Plant propagation: principles and practices. Upper Saddle River, NJ: Prentice Hall. 2002; 253-257.

32. Sachs T. Auxin's role as an example of the mechanisms of shoot/root relation. Plant Soil. 2005;268: 13-19.
33. Husen A, Pal M. Metabolic changes during adventitious root primordium development in Tectona grandis Linn. (teak) cuttings as affected by age of donor plants and auxin (IBA and NAA) treatment. New For. 2007;33:309–323.
34. Polat AA. Effect of IBA on rooting mulberry cuttings. Acta Hort. 2008;774(774): 351-354.
35. Kalyoncu IH, Ersoy NY, Aydyn M. Effect of humidity level and IBA dose application on the softwood top cuttings of white mulberry (Morus nigra L.,) types. Africa J. Biotechnol. 2009;8(16): 3754-3760.
36. Chumpookam J, Arunjit P, Theanhom AA. Effect of IBA and NAA on rooting and axillary shoot outgrowth of 'Chiangmai 60' mulberry (Morus alba L.) stem cutting. Khon Kaen Agriculture Journal, 3: 162-167.