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Study on the Effect of Auxin in *In vitro* Root Regeneration and Survival of *Dendrobium* cv. Sonia Earsakul

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

The investigation was carried out during 2017-2019 at the Tissue Culture Laboratory, Department of Horticulture, NU: SASRD, Nagaland to study the influence of different auxins in varied doses on *in vitro* root regeneration of *Dendrobium* cv. Sonia Earsakul. The different treatments used are IBA, NAA and IAA @ 0, 0.5, 1, 1.5 and 2 mgL^{-1} respectively. The study revealed that among the different treatments IBA @1 mg/l resulted in the longest root length (5.97 cm), longest shoot length (7.8 cm), highest survivability during primary hardening (34.30%) and secondary hardening (94.29%).

Keywords: Dendrobium cv. Sonia earsakul; IBA; NAA; IAA.

1. INTRODUCTION

Dendrobium the orchidaceae family, with about 1,100 species, of which at least 300 have been cultivated make *Dendrobium* the second largest orchid genus and most commonly encountered orchids in retail trade [1]. This mesmerizing *Dendrobium* is well known in the orchidaceae family for their complex fabricated and long lasting colourful flowers and are highly valued in the flower industry as potted plant and cut flower for their marvelously long lasting flower. *Dendrobium*s are sturdy orchids that can be added as a focal point to any room. *Dendrobium* is well known in the orchidaceae family for their complex fabricated and long lasting colourful

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flowers. Different characteristics of *Dendrobium* such as rapid growth, easiness of plantlet regeneration, beauty of the flower, and year round production in control flowering and long lasting of the flower stalk are very advantageous of this genus [2]. Orchids have a low rate of multiplication under natural or green house conditions. It thus requires a rapid and efficient micropropagation protocol for obtaining true-to-type regenerants without detriment to the survival of mother plant to meet the demand and saving its populations from getting rarer in nature. The development of protocols for extensive and clonal multiplication of highly priced varieties of *Dendrobium* serves as a foundation for commercial scale propagation. Micropropagation of orchids is the most frequently used convenient technique for their exploitation as a major trade in developed countries [3]. *In vitro* propagation of orchid offers an opportunity for the selection of various desirable traits and produces high quality and uniform plantlets throughout the year under disease free conditionsregardless of the seasons and weathers [4]. Orchids can be rapidly propagated through tissue culture techniques by using shoot tips [5]. Plant growth regulators are essential part for *in vitro* regeneration of crop grown in artificial medium. Optimisation of plant growth regulators is prerequisite for any *in vitro* micropropagtion work. The development of protocols for extensive and clonal multiplication of highly priced varieties of *Dendrobium* serves as a foundation for commercial scale propagation. Thus, the present study was undertaken to study the effect of some auxins on root regeneration and performance of rooted *in vitro* induced shoots during hardening.

2. MATERIALS AND METHODS

The study was carried out at the Tissue Culture Laboratory, Department of Horticulture, School of Agricultural Sciences and Rural Development, Nagaland University, Medziphema campus during the year 2017-2019. The experiment was laid out in Completely Randomized Design with 13 different treatments to study the effect of different auxins on the in vitro root regeneration and percentage of surviving of *Dendrobium* cv. Sonia Earsakul. The different treatments used in MS media for the study are IBA @ 0, 0.5, 1, 1.5, 2 mgL^{-1}, NAA @ 0, 0.5, 1, 1.5, 2 mgL^{-1} and IAA @ 0, 0.5, 1, 1.5, 2 mgL^{-1}. Rootless shoot developed from shooting media containing BAP @ 2 mgL^{-1} and NAA @ 0.5 mgL^{-1} were used to study the *in vitro* root regeneration of *Dendrobium* cv. Sonia Earsakul. Rootless developed shoot were inoculated in culture bottle containing different rooting media as per the experiment under control environment. The well developed rooted plantlets were hardened in the potting media containing coco peat and agricultural perlite in 4:1 ratio in natural environment in polyhouse. The different parameters recorded were length of roots, length of shoot, percentage of survival during primary and secondary hardening.

3. RESULTS AND DISCUSSION

3.1 Length of Root

Analysis of data given in Table 1 Fig. 1, revealed the effect of various plant growth regulators in different levels on the length of root of *Dendrobium* cv. Sonia Earsakul for *in vitro* root regeneration. During the year 2017-18, the longest length of root (6.05 cm) was recorded in IBA @ 1 mL^{-1} followed by IBA @0.5 mgL^{-1} (0.93 cm) and the shortest length of root (4.78 cm) was recorded in control. Similarly, in the subsequent year 2018-19, the longest length of root (5.90 cm) was recorded in IBA @ 1 mgL^{-1}followed by IBA @ 1.5 mgL^{-1} (5.85 cm) which is at par with NAA @ 1 mgL^{-1} (5.83 cm) and the shortest length of root (4.63 cm) was recorded in control. Pooled data analysis of both the years revealed significant difference in the length of the root. The longest length of root (5.97 cm) was recorded in IBA @ 1 mgL^{-1} which is followed by IBA @ 0.5 mgL^{-1}(5.88 cm) which is at par with IBA @ 1.5 mgL^{-1}and NAA @ 1 mgL^{-1} (5.86 cm each). The shortest length of root (4.71 cm) was recorded in control. Each species prefer particular hormones concentrations reported by Dunwell [6]. The current findings are in conformity with the experimental finding of researchers Nongdam and Tikendra [7] where they reported that IBA gave better result in root regeneration as compared to NAA and IAA. These results are in conformity with the findings of Aktar et al. [8] in *Dendrobium* orchid, Panathula et al. [9] in *Centella asiatica* (L.) and Nongdam and Tikendra [7] in *Dendrobium chrysotoxum.*

3.2 Length of Shoot

The data on the length of shoot as influenced by the plant growth regulators are presented in Table 1 and Fig. 2. The longest length of shoot (6.97 cm) during the year 2017-18 were recorded in IBA @ 1 mgL^{-1} followed by IBA @ 1.5 mgL^{-1}(6.82 cm) which is at par with NAA @ 1 mgL^{-1}(6.80 cm) and the shortest length of shoot 3.93 were recorded in control. Similar results were obtained in the subsequent year 2018-19, the longest length of shoot (6.90 cm) was recorded in IBA @ 1 mgL^{-1}followed by IBA @1.5 mgL^{-1}(6.82 cm) which is at par with NAA @ 1 mgL^{-1} (6.80 cm) and the shortest length of shoot (3.93 cm) was recorded in control. Pooled data analysis revealed that the longest length of shoot (6.93 cm) was recorded in IBA @ 1 mgL^{-1}followed by NAA @ 1 mgL^{-1}(6.83 m) which is at par with IBA @ 1.5 mgL^{-1}and IBA @ 2 mgL^{-1}(6.82 cm and 6.78 cm respectively). *In vitro* multiplication of shoot was carried out at high concentration of cytokinin which inhibited the development of shoot, to induce root lower cytokinin/auxin ratio is favourable. The reason for getting such length of shoot might be due to the fact that better root formation results in better uptake of nutrient from the media which lead to better growth of the shoot. The current experimental findings are in line with the findings of Panathula et al. [9] in *Centella asiatica* (L), Aktar et al. [8] in *Dendrobium* orchid.

Table 1. Influence of auxins on length of root and shoot in *in vitro* regeneration *of Dendrobium* cv. Sonia Earsakul

Treatment	Length of root			Length of shoot		
	2017-18	2018-19	Pooled	2017-18	2018-19	Pooled
T_1(MS)	4.78^{d}	4.63^{e}	4.71^{g}	3.93^{h}	4.00^{f}	3.97^{h}
T_2(MS + 0.5 mg/l IBA)	5.93^{ab}	5.82^{ab}	5.88^{ab}	4.93^{f}	4.80^{e}	4.87^{f}
T_3(MS + 1 mg/l IBA)	6.05^{a}	5.90^{a}	5.97^{a}	6.97^{a}	6.90^{a}	6.93^{a}
T_4(MS + 1.5 mg/l IBA)	5.87^{abc}	5.85^{ab}	5.86^{ab}	6.82^{ab}	6.82^{ab}	6.82^{ab}
T_5(MS + 2 mg/l IBA)	5.83^{abc}	5.78^{ab}	5.81^{bc}	6.77^{abc}	6.78^{ab}	6.78^{ab}
T_6(MS + 0.5 mg/l NAA)	5.85^{abc}	5.78^{ab}	5.82^{abc}	4.87^{f}	4.93^{e}	4.90^{f}
T_7(MS + 1 mg/l NAA)	5.90^{abc}	5.83^{ab}	5.86^{ab}	6.80^{ab}	6.86^{a}	6.83^{ab}
T_8(MS + 1.5 mg/l NAA)	5.75^{bc}	5.68^{b}	5.72^{bc}	6.65^{bc}	6.72^{ab}	6.68^{bc}
T_9(MS + 2 mg/l NAA)	5.60^{c}	5.45^{c}	5.53^{de}	6.35^{e}	6.34^{d}	6.34^{e}
T_{10}(MS + 0.5 mg/l 1AA)	5.62^{c}	5.70^{b}	5.66^{cd}	4.58^{g}	4.77^{e}	4.68^{g}
T_{11}(MS + 1 mg/l 1AA)	5.80^{abc}	5.75^{ab}	5.78^{bc}	6.52^{cd}	6.64^{bc}	6.58^{cd}
T_{12}(MS + 1.5 mg/l 1AA)	5.78^{abc}	5.17^{d}	5.48^{ef}	6.48^{de}	6.46^{cd}	6.47^{de}
T_{13}(MS + 2 mg/l 1AA)	5.63^{bc}	5.06^{d}	5.35^{f}	6.47^{de}	6.37^{d}	6.42^{de}
SEm±	*0.08*	*0.05*	*0.05*	*0.09*	*0.06*	*0.05*
CD (*p*=.05)	*0.24*	*0.16*	*0.14*	*0.25*	*0.19*	*0.15*

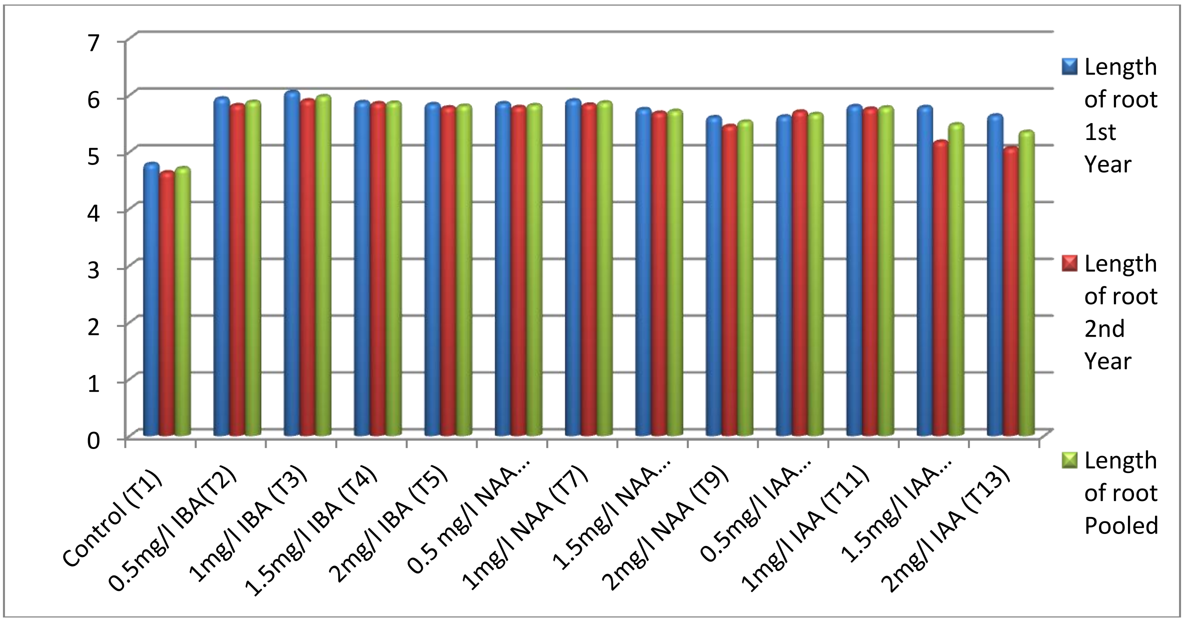

Fig. 1. Influence of different auxins on length of root

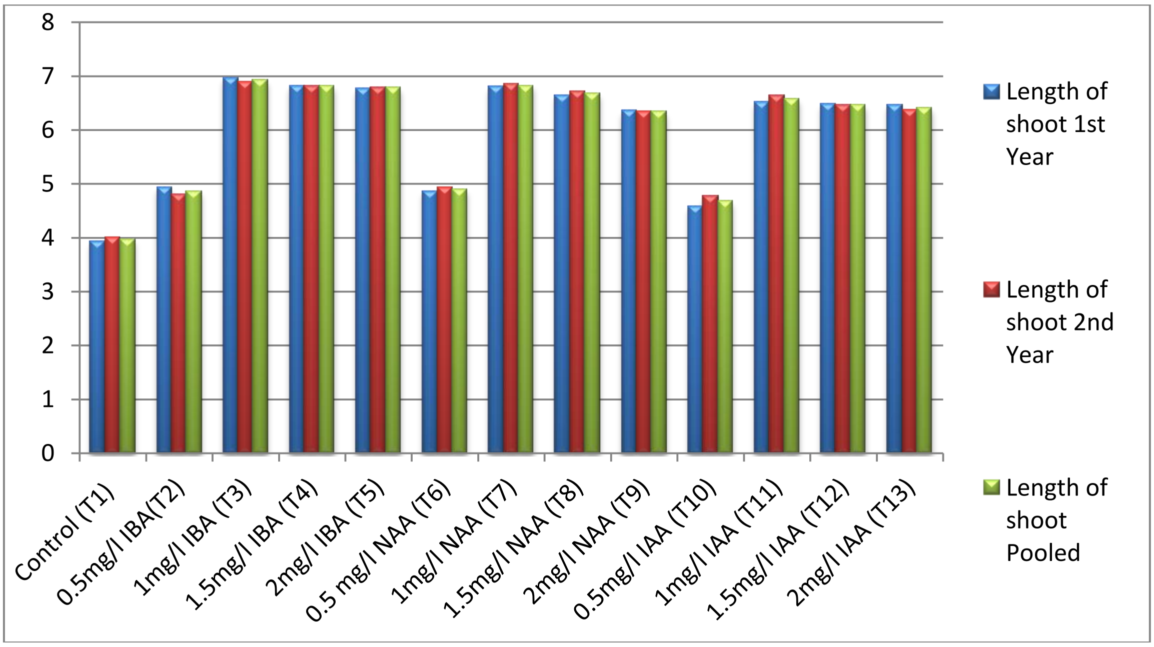

Fig. 2. Influence of different auxins on length of shoot

Table 2. Influence of auxins on survival percentage of rooted plantlets during of *Dendrobium* cv. Sonia Earsakul

Treatment	Primary hardening			Secondary hardening		
	2017-18	2018-19	Pooled	2017-18	2018-19	Pooled
T_1(MS)	27.33^{g}	26.87^{g}	27.10^{f}	82.20^{f}	83.77^{f}	82.99^{g}
T_2(MS + 0.5 mg/l IBA)	31.00^{de}	30.34^{de}	30.67^{c}	89.77^{de}	92.16^{bc}	90.97^{c}
T_3(MS + 1 mg/l IBA)	34.69^{a}	33.91^{a}	34.30^{a}	94.29^{a}	96.37^{a}	95.33^{a}
T_4(MS + 1.5 mg/l IBA)	33.57^{ab}	32.27^{bc}	32.92^{b}	92.54^{ab}	95.75^{a}	94.15^{a}
T_5(MS + 2 mg/l IBA)	32.45^{bc}	32.03^{bc}	32.24^{b}	91.92^{bc}	93.26^{b}	92.59^{b}
T_6(MS + 0.5 mg/l NAA)	29.67^{ef}	29.37^{ef}	29.52^{de}	88.12^{e}	89.62^{d}	88.87^{de}
T_7(MS + 1 mg/l NAA)	33.18^{b}	32.92^{ab}	33.05^{b}	89.60^{d}	92.12^{bc}	90.86^{c}
T_8(MS + 1.5 mg/l NAA)	32.83^{bc}	32.00^{bc}	32.42^{b}	91.37^{bcd}	91.14^{bcd}	91.26^{bc}
T_9(MS + 2 mg/l NAA)	31.00^{de}	31.00^{cd}	31.00^{c}	88.84^{e}	91.04^{cd}	89.94^{cde}
T_{10}(MS + 0.5 mg/l 1AA)	28.50^{fg}	28.70^{f}	28.60^{e}	84.29^{f}	86.28^{e}	85.29^{f}
T_{11}(MS + 1 mg/l 1AA)	32.77^{bc}	32.42^{ab}	32.59^{b}	88.40^{e}	89.11^{d}	88.76^{de}
T_{12}(MS + 1.5 mg/l 1AA)	31.60^{cd}	30.27^{de}	30.93^{c}	89.41^{d}	91.12^{bcd}	90.27^{cd}
T_{13}(MS + 2 mg/l 1AA)	30.97^{de}	29.63^{de}	30.30^{cd}	88.08^{e}	88.91^{d}	88.49^{e}
SEm±	0.46	0.50	0.34	0.72	0.67	0.49
CD (p=0.5)	1.33	1.45	0.96	2.09	1.96	1.40

3.3 Percentage of Survival during Primary Hardening

The data pertaining to percentage of survival during the primary hardening as influenced by different auxins in various levels are presented in Table 2 During the first year study 2017-18, results revealed significant difference, application of IBA @ 1 mgL^{-1}resulted in the highest percentage of survival (34.69 %) followed by IBA @ 1.5 mgL^{-1} (33.57%) and the least percentage of survival (27.33 %) was recorded in control. In the subsequent year study 2018-19, IBA @ 1 mgL^{-1}resulted in the highest percentage of surviving (33.91%) followed by IAA @ 1 mgL^{-1} (32.42%) and the least percentage of survival (26.87%) was recorded in control. Further analysis of pooled data resulted in significant difference in the percentage of survival during primary hardening. MS media supplemented with IBA @ 1 mgL^{-1}resulted in highest percentage of survival (34.30%) followed by NAA @ 1 mgL^{-1}(33.05%) which is at par with IBA @ 1.5 mgL^{-1} (32.92%), IAA @ 1 mgL^{-1}(32.59%), NAA @ 1.5

mgL^{-1} (32.42%) and IBA @ 2 mgL^{-1} (32.24%) whereas the least percentage of survival (27.10%) was recorded in control.

3.4 Percentage of Survival during Secondary Hardening

Table 2 depict the results of the influence of auxins on the percentage of survival on the hardening of *Dendrobium* cv. Sonia Earsakul. During the first year (2017-18), the highest percentage of survival (94.29%) was recorded with IBA @ 1 mgL^{-1} which is followed by IBA @ 1.5 mgL^{-1} (92.54%) which is at par with IBA @ 2mgL^{-1} (91.92%) and the least percentage of survival (82.20%) was recorded in control. In the second year (2018-19), the highest percentage of survival (96.37%) was recorded with IBA @ 1 mgL^{-1}which is at par with IBA @ 1.5 mgL^{-1} (95.75%) followed by IBA @ 2 mgL^{-1} (93.26%) and the least percentage of survival was recorded in control (83.77). Pooled data analysis of both the years showed that the highest percentage of survival (95.33%) was recorded with IBA @ 1 mgL^{-1} which is at par with IBA @ 1.5 mgL^{-1}(94.15%) followed by IBA @ 2 mgL^{-1} (92.59%)and the least percentage survival was recorded in control (82.99%). During the primary hardening, the overall percentage of survival was less but during the secondary hardening the rate of survival increased. During the primary hardening the tender seedlings from *in vitro* culture bottles were acclimatized and hardened in natural environment. Survival percentage in primary hardening was less which might be due to the sudden shock, tenderness of plants, adverse natural environment, susceptibility to pests, diseases and other factors. However, the percentage of survival was quite satisfactory in the secondary hardening, as the plant were getting sturdy and adapted to the natural environment. In both the primary and secondary hardenings the best percentage of survival was recorded in treatment containing IBA @ 1 mgL^{-1}, which might be due to proper root development in that treatment where *in vitro* plants were growing in culture bottle comparing to other treatments. Plantlets with proper root development will establish easily in the natural environment during hardening and thus resulting in proper growth and development. These findings are in accordance with the findings of Priyanka et al. [10] in *Dendrobium* sp. where 85% of plantlets developed from media augmented with IBA @ IBA 1 mgL^{-1} survived in shade house, Panathula et al. [9] in *Centella asiatica* (L.) reported that IBA @1 mgL^{-1} resulted in 85% survival during hardening, Anbazhagan et al. [11] in *Musa* sp. and Panathula *et al*. [9] in *Centella asiatica* (L.).

4. CONCLUSION

In conclusion, the results of the present study indicated that for root length, length of shoot and percentage of survival during primary and secondary was superior with the supplementation of MS media with IBA @ 1 mgL^{-1}in *Dendrobium* cv. Sonia Earsakul.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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