Effects of Different Hormones (NAA and BAP) Combination on Anthurium andraeanum Linden Ex Andre

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Abstract

The plantlets of *Anthurium andraeanum* Linden ex Andre were obtained from Mingalardon Orchid Garden, Mingalardon Township and Yangon Region. The plantlets of *Anthurium andraeanum* Linden ex Andre were grown on MS (Murashige and Skoog, 1962). The effect of various concentrations of plant growth hormones combination NAA (naphthalene acetic acid) + BAP (6-benzylaminopurine) were used. The maximum growth and development of plantlets was obtained from NAA (0.5 mg/l) + BAP (1.0 mg/l) in all treatments. The growth of plantlets was recorded by color photographs. This experiment was carried out in the tissue culture laboratory, Department of Botany, Dagon University from December 2022 to June 2023.

Introduction

Anthurium andraeanum Linden ex Andre belong to the Araceae family, which consists of 108 genera and approximately 3750 species. Anthuriums are primarily a tropical family, found in a temperature range from 16°C to 30°C. Araceae are particularly used as ornamentals (Grayum, 1990). Anthurium andraeanum Linden ex Andre is a modified leaf (spathe), bearing numerous small flowers on a pencil – like protrusion (spadix) and has a vase life of 14 – 28 days. Different variant of Anthurium are grown for both foliage and brightly coloured attractive waxy spathes (George, 1951).

Anthurium andraeanum Linden ex Andre is commonly grown for cut flowers and sometimes adaptable to pot culture. Anthurium's global market among all tropical cut flowers is the second biggest, only next to Orchids (Reisch, 1998; Agampodi and Jayawardena, 2007; Gantait and Mandal, 2010; Junqueira and Peetz, 2012). Bangladesh has a scope of large scale production of Anthurium and other tropical ornamental plants through micro-propagation method for quality production to meet the demand on local and international market (Ullah, 1995).

Micropropagation of Anthurium has been achieved with various explants. The tissue culture of Anthurium was first reported by Pierik *et al.* (1974). Most of the cultivated Anthurium are found as a rare species. Therefore, to get true to type plant, micro-propagation is the only means presently involved in exploitation as a major trade worldwide (Healy *et al.*, 1980; Sagawa and Kunisaki, 1982; Goh *et al.*, 1992).

This is because commercially mass propagation is possible by production millions of plants using tissue culture techniques (Lim - Ho *et al.*, 1985). Auxins are often most effective when combined with cytokinins.

The present studied to produce and multiply the virus free plants in a short period of time, to study the growth and development of *Anthurium andraeanum*

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Linden ex Andre. plantlets cultured on various hormones and to study shoot multiplication and root induction of *Anthurium andraeanum* Linden ex Andre.

Materials and Methods

Source of plant material

The plantlets of *Anthurium andraeanum* Linden ex Andre were obtained from Mingalardon Orchid Garden, Mingalardon Township, Yangon Region. The plantlets were maintained on MS medium.



Figure (1) Initial stage of Anthurium andraeanum Linden ex Andre explants

Identification of Anthurium andraeanum Linden ex Andre

The morphological studies were made from the collected specimen by using available literature such as Hundley and Chit Ko Ko (1987) and Datta (1970).

Media preparation

According to Murashige and Skoog (1962), method of 1L medium preparation applied in the present study is as follows;

- 50 ml of stock solution were poured into 1000 ml beaker.
- 30 g/l of sucrose were added and stirred.
- The required amount of distilled water was added to fulfill the 1L media concentration.
- The pH value of the media was measured and adjusted 5.8 with NaOH and HCl.
- The solution was heated and stirred continuously until boiled.
- Agar powder (8 g/l) were slowly added and stirred to dissolve the agar and then the media became amber colored.
- 20 ml of the medium was poured into 150 ml size culture bottles.
- The culture bottles were covered and sterilized by autoclaving at 121°C (1.5 kg/cm²) pressure for 15 mins.
- Then the bottles were cooled and used.

Incubation

All cultured bottles were incubated in culture room at the temperature of 24 ± 1 °C. Light was supplied by 4 feet fluorescent tubes and photoperiod of 16/8 hours (light/dark).

Data Collection

Data collections were done in this experiment. Each treatment has three replicated.

Shoot tips were culture in prepared media

MS basal solid medium supplemented with different hormones combination of NAA (0.5 mg/l, 1.0 mg/l, 2.0 mg/l and 4.0 mg/l) + BAP (0.5 mg/l, 1.0 mg/l, 2.0 mg/l and 4.0 mg/l

Inoculation

Growth of plantlets as shoots and leaves formation

Data collection and measurement Number of shoots Length of shoots Number of leaves Length of leave

Word diagram of experiments (1), (2), (3) and (4)

Table 1. Different hormones combination of NAA + BAP supplemented to MS solid medium

Treatment	Basal Medium	NAA+BAP Concentration
T ₀ (control)	MS	-
T_1	MS	0.5 mg/l + 0.5 mg/l
T ₂	MS	0.5 mg/l + 1.0 mg/l
T ₃	MS	0.5 mg/l + 2.0 mg/l
T ₄	MS	0.5 mg/l + 4.0 mg/l

Table 2. Different hormones combination of NAA + BAP supplemented to MS solid medium

Treatment	Basal Medium	NAA+BAP Concentration
T ₀ (control)	MS	-
T ₁	MS	1.0 mg/l + 0.5 mg/l
T ₂	MS	1.0 mg/l + 1.0 mg/l
T ₃	MS	1.0 mg/l + 2.0 mg/l
T ₄	MS	1.0 mg/l + 4.0 mg/l

Table 3. Different hormones combination of NAA + BAP supplemented to MS solid medium

Treatment	Basal Medium	NAA+BAP Concentration
T ₀ (control)	MS	-
T ₁	MS	2.0 mg/l + 0.5 mg/l
T ₂	MS	2.0 mg/l + 1.0 mg/l
T ₃	MS	2.0 mg/l + 2.0 mg/l
T ₄	MS	2.0 mg/l + 4.0 mg/l

Table 4. Different hormones combination of NAA + BAP supplemented to MS solid medium

Treatment	Basal Medium	NAA+BAP Concentration
T ₀ (control)	MS	-
T ₁	MS	4.0 mg/l + 0.5 mg/l
T_2	MS	4.0 mg/l + 1.0 mg/l
T ₃	MS	4.0 mg/l + 2.0 mg/l
T ₄	MS	4.0 mg/l + 4.0 mg/l

Results

Morphological Characters of Anthurium andraeanum Linden ex Andre

Scientific Name - Anthurium andraeanum Linden ex Andre

Family – Araceae

Myanmar Name – Pain

- Habit; Perennial herb.
- Leaves; Simple, dark-green, apex acuminate, cordate base, petiole long.
- Inflorescences; Spadix, subtended by a spathe.
- Flowers; Bisexual, red color, pedicels cylindrical long.



Figure (2) Habit

Effect of NAA (0.5 mg/l) and Different Combination of BAP in MS Solid Medium

Plantlets of *Anthurium andraeanum* Linden ex Andre were cultured in MS solid medium containing different hormones combination of NAA (0.5 mg/l) + BAP (0.5 mg/l, 1.0 mg/l, 2.0 mg/l and 4.0 mg/l). Each plantlets was cultured in five treatments. Three replications were carried out in each treatment. The results are showed in Table (5) and Figure (3).

The results of T_0 (without hormones), the number of shoots had 1.51 and the length of shoots had 0.15 cm. The number of leaves had 1.33 and the length of leaves had 0.24 cm.

The results of T_1 (0.5 mg/l + 0.5 mg/l), the number of shoots had 1.72 and the length of shoots had 0.38 cm. The number of leaves had 1.42 and the length of leaves had 0.43 cm.

The results of T_2 (0.5 mg/l + 1.0 mg/), the number of shoots had 2.43 and the length of shoots had 0.44 cm. The number of leaves had 2.22 and the length of leaves had 0.51 cm.

The results of T_3 (0.5 mg/l + 2.0 mg/l), the number of shoots had 1.78 and the length of shoots had 0.28 cm. The number of leaves had 1.39 and the length of leaves had 0.42 cm.

The results of T_4 (0.5 mg/l + 4.0 mg/l), the number of shoots had 1.54 and the length of shoots had 0.21 cm. The number of leaves had 1.37 and the length of leaves had 0.39 cm.

In this experiment, NAA 0.5 mg/l and different combination of BAP 1.0 mg/l is best result followed by 4.0 mg/l, 0.5 mg/l and 2.0 mg/l. However, the control had the least growth parameter was observed from this experiment.

Table 5. Growth parameters of plantlets in various concentration of plant growth hormones combination NAA (0.5 mg/l) + BAP (0.5 mg/l, 1.0 mg/l, 2.0 mg/l and 4.0 mg/l) in Anthurium andraeanum Linden ex Andre

Treatment	MS media with different combination of NAA+BAP	Number of shoots	Length of shoots	Number of leaves	Length of leaves
T_0	0 mg/l	1.51	0.15	1.33	0.24
T_1	0.5 mg/l + 0.5 mg/l	1.72	0.38	1.42	0.43
T_2	0.5mg/l+1.0mg/l	2.43	0.44	2.22	0.51
T ₃	0.5mg/l+2.0mg/l	1.78	0.28	1.39	0.42
T_4	0.5mg/l+4.0mg/l	1.54	0.21	1.37	0.39



Figure (3) Comparison of *Anthurium andraeanum* Linden ex Andre growth and development on various concentration of hormones combination NAA (0.5 mg/l) + BAP (0.5 mg/l, 1.0 mg/l, 2.0 mg/l and 4.0 mg/l) in 8 weeks old culture

Effect of NAA (1.0 mg/l) and Different Combination of BAP in MS Solid Medium

Plantlets of *Anthurium andraeanum* Linden ex Andre were cultured in MS solid medium containing different hormones combination of NAA (1.0 mg/l) + BAP (0.5 mg/l, 1.0 mg/l, 2.0 mg/l) and 4.0 mg/l). Each plantlets having $1 \pm 1 \text{ mg}$ in fresh weight was cultured in five treatments. Three replications were carried out in each treatment. The results are showed in Table (6) and Figure (4).

The results of T_0 (without hormones), the number of shoots had 1.51 and the length of shoots had 0.15 cm. The number of leaves had 1.33 and the length of leaves had 0.24 cm.

The results of T_1 (1.0 mg/l + 0.5 mg/l), the number of shoots had 1.53 and the length of shoots had 0.24 cm. The number of leaves had 1.35 and the length of leaves had 0.38 cm.

The results of T_2 (1.0 mg/l + 1.0 mg/l), the number of shoots had 1.84 and the length of shoots had 0.42 cm. The number of leaves had 1.58 and the length of leaves had 0.62 cm.

The results of T_3 (1.0 mg/l + 2.0 mg/l), the number of shoots had 1.71 and the length of shoots had 0.35 cm. The number of leaves had 1.39 and the length of leaves had 0.54 cm.

The results of T_4 (1.0 mg/l + 4.0 mg/l), the number of shoots had 1.58 and the length of shoots had 0.29 cm. The number of leaves had 1.36 and the length of leaves had 0.43 cm.

In this experiment, NAA 1.0 mg/l and different combination of BAP 1.0 mg/l is best result followed by 0.5 mg/l, 4.0 mg/l and 2.0 mg/l. However, the control had the least growth parameter was observed from this experiment.

Table 6. Growth parameters of plantlets in various concentration of plant growth hormones combination NAA (1.0 mg/l) + BAP (0.5 mg/l, 1.0 mg/l, 2.0 mg/l and 4.0 mg/l)in *Anthurium andraeanum* Linden ex Andre

Treatment	MS media with different combination of NAA+BAP	Number of shoots	Length of shoots	Number of leaves	Length of leaves
T_0	0 g/l	1.51	0.15	1.33	0.24
T_1	1.0mg/l+0.5mg/l	1.53	0.24	1.35	0.38
T_2	1.0mg/l+1.0mg/l	1.84	0.42	1.58	0.62
T ₃	1.0mg/l+2.0mg/l	1.71	0.35	1.39	0.54
T ₄	1.0mg/l+4.0mg/l	1.58	0.29	1.36	0.43



Figure (4) Comparison of *Anthurium andraeanum* Linden ex Andre growth and development on various concentration of hormones combination NAA (1.0 mg/l) + BAP (0.5 mg/l, 1.0 mg/l, 2.0 mg/l and 4.0 mg/l) in 8 weeks old culture

Effect of NAA (2.0 mg/l) and Different Combination of BAP in MS Solid Medium

Plantlets of *Anthurium andraeanum* Linden ex Andre were cultured in MS solid medium containing different hormones combination of NAA (2.0 mg/l) + BAP (0.5 mg/l, 1.0 mg/l, 2.0 mg/l) and 4.0 mg/l). Each plantlets having 1 ± 1 mg in fresh weight was cultured in five treatments. Three replications were carried out in each treatment. The results are showed in Table (7) and Figure (5).

The results of T_0 (without hormones), the number of shoots had 1.51 and the length of shoots had 0.15 cm. The number of leaves had 1.33 and the length of leaves had 0.24 cm.

The results of T_1 (2.0 mg/l + 0.5 mg/l), the number of shoots had 1.57 and the length of shoots had 0.23 cm. The number of leaves had 1.36 and the length of leaves had 0.34 cm.

The results of T_2 (2.0 mg/l + 1.0 mg/l), the number of shoots had 1.63 and the length of shoots had 0.37 cm. The number of leaves had 1.69 and the length of leaves had 0.56 cm.

The results of T_3 (2.0 mg/l + 2.0 mg/l), the number of shoots had 1.59 and the length of shoots had 0.28 cm. The number of leaves had 1.38 and the length of leaves had 0.43 cm.

The results of T_4 (2.0 mg/l + 4.0 mg/l), the number of shoots had 1.54 and the length of shoots had 0.19 cm. The number of leaves had 1.35 and the length of leaves had 0.29 cm.

In this experiment, NAA 2.0 mg/l and different combination of BAP 1.0 mg/l is best result followed by 2.0 mg/l, 4.0 mg/l and 0.5 mg/l. However, the control had the least growth parameter was observed from this experiment.

Table 7. Mean value of the number of shoots, leaves and the length of shoots, leaves of *Anthurium andraeanum* Linden ex Andre cultured on MS solid medium with different hormones combination of NAA (2.0 mg/l) + BAP (0.5, 1.0, 2.0 and 4.0 mg/l)

Treatment	MS media with different combination of NAA+BAP	Number of shoots	Length of shoots	Number of leaves	Length of leaves
T_0	0 mg/l	1.51	0.15	1.33	0.24
T_1	2.0mg/l+0.5mg/l	1.57	0.23	1.36	0.34
T_2	2.0mg/l+1.0mg/l	1.63	0.37	1.69	0.56
T ₃	2.0mg/l+2.0mg/l	1.59	0.28	1.38	0.43
T ₄	2.0mg/l+4.0mg/l	1.54	0.19	1.35	0.29



Figure (5) Comparison of *Anthurium andraeanum* Linden ex Andre growth and development on various concentration of hormones combination NAA (2.0 mg/l) + BAP (0.5 mg/l, 1.0 mg/l, 2.0 mg/l and 4.0 mg/l) in 8 weeks old culture

Effect of NAA (4.0 mg/l) and Different Combination of BAP in MS Solid Medium

Plantlets of *Anthurium andraeanum* Linden ex Andre were cultured in MS solid medium containing different hormones combination of NAA (4.0 mg/l) + BAP (0.5 mg/l, 1.0 mg/l, 2.0 mg/l) and 4.0 mg/l). Each plantlets having $1 \pm 1 \text{ mg}$ in fresh weight was cultured in five treatments. Three replications were carried out in each treatment. The results are showed in Table (8) and Figure (6).

The results of T_0 (without hormones), the number of shoots had 1.51 and the length of shoots had 0.15 cm. The number of leaves had 1.33 and the length of leaves had 0.24 cm.

The results of T_1 (4.0 mg/l + 0.5 mg/l), the number of shoots had 1.89 and the length of shoots had 0.34 cm. The number of leaves had 1.62 and the length of leaves had 0.49 cm.

The results of T_2 (4.0 mg/l + 1.0 mg/l), the number of shoots had 1.63 and the length of shoots had 0.29 cm. The number of leaves had 1.42 and the length of leaves had 0.36 cm.

The results of T_3 (4.0 mg/l + 2.0 mg/l), the number of shoots had 1.56 and the length of shoots had 0.23 cm. The number of leaves had 1.39 and the length of leaves had 0.31 cm.

The results of T_4 (4.0 mg/l + 4.0 mg/l), the number of shoots had 1.53 and the length of shoots had 0.19 cm. The number of leaves had 1.37 and the length of leaves had 0.26 cm.

In this experiment, NAA 4.0 mg/l and different combination of BAP 1.0 mg/l is best result followed by 2.0 mg/l, 4.0 mg/l and 0.5 mg/l. However, the control had the least growth parameter was observed from this experiment.

Table 8. Mean value of the number of shoots, leaves and the length of shoots, leaves of *Anthurium andraeanum* Linden ex Andre cultured on MS solid medium with different hormones combination of NAA (4.0 mg/l) + BAP (0.5, 1.0, 2.0 and 4.0 mg/l)

Treatment	MS media with different combination of NAA+BAP	Number of shoots	Length of shoots	Number of leaves	Length of leaves
T_0	0 mg/l	1.51	0.15	1.33	0.24
T_1	4.0mg/l+0.5mg/l	1.89	0.34	1.62	0.49
T_2	4.0mg/l+1.0mg/l	1.63	0.29	1.42	0.36
T ₃	4.0mg/l+2.0mg/l	1.56	0.23	1.39	0.31
T ₄	4.0mg/l+4.0mg/l	1.53	0.19	1.37	0.26



Figure (6) Comparison of *Anthurium andraeanum* Linden ex Andre. growth and development on various concentration of hormones combination NAA (4.0 mg/l) + BAP (0.5 mg/l, 1.0 mg/l, 2.0 mg/l and 4.0 mg/l) in 8 weeks old culture

Discussion and Conclusion

The morphological characters of *Anthurium andraeanum* Linden ex Andre are perennial herb, leaves simple and acuminate, inflorescence spadix, flower acuminate. The finding agreed with Chit Ko Ko (1987) and Datta (1970). In the present study, The plantlets of *Anthurium andraeanum* Linden ex Andre were cultured on MS (Murashige and Skoog, 1962) medium with different hormones combination of NAA (0.5, 1.0, 2.0 and 4.0 mg/l) each with BAP (0.5, 1.0, 2.0 and 4.0 mg/l) respectively.

After 2 months culture, the highest growth value was observed T_2 NAA (0.5 mg/l) + BAP (1.0 mg/l) in experiment 1, T_2 NAA (1.0 mg/l) + BAP (1.0 mg/l) in experiment 2, T_2 NAA (2.0 mg/l) + BAP (1.0 mg/l) in experiment 3 and T_1 NAA (4.0 mg/l) + BAP (0.5 mg/l) in experiment 4. This result agreed with ISLAM *et al.*, 2010 who got the best growth at NAA (1.0 mg/l) + BAP (1.0 mg/l) of *Anthurium andraeanum* cv. Nitta.

In conclusion, in all experiments, combination of hormones NAA (0.5 mg/l) + BAP (1.0 mg/l) is the best growth and development of *Anthurium andraeanum* Linden ex Andre. *Anthurium andraeanum* Linden ex Andre is the second great demand in commercial market in worldwide and so it need to conserve with plant tissue culture technique.

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References

- Agampodi, V.A. and B.M. Jayawardena, 2007. Effect of coconut water in extending the vase life of *Anthurium* cut flower variety wild pink. *Tropic. Agric. Res.*, 19: 202-209.
- Datta, S.C. (1970). A Handbook of Systematic Botany. University of Calcutta.
- Grayum, M.H. 1990. Evolution and phylogeny of the Araceae. Annals of the Missouri Botanical Gardens 77, 629-697.
- George, H. M. L. 1951. Taxonomy of vascular plants. Mohan Primlani, Oxford and IBH Publishing Co.66 Janpath, New Delhi. pp: 398-400.
- Gantiat, S. and N. Mandal, 2010. Tissue culture of *Anthurium andraeanum*: a significant review and future prospective. *Int. J. Botany*, 6: 207-219.
- Goh, C. J. A. A. Sim and G. Lim. 1992. Mycorrhizal associations in some tropical orchids. Lindleyana, 7(1): 13-17. [Cited from Hort. Abstr., 59(5): 3948, 1994].
- Healy, P. L., J. D. Michaud and J. Ariditti. (1980). Morphometry of orchid seeds. III. Native California and related species of *Goodeyera*, *Piperia*, *Plantanthera* and Spiranthes. *Amer.* J. Bot., 97: 508-518.
- Hundly, H.G and Chit Ko Ko. (1987). List of Trees, Shrubs, Herbs and Principle Climbers of Myanmar. Government Printing Press, Yangon, Myanmar.
- S.A. ISLAM, (2010). *In Vitro* Regeneration of *Anthurium andraeanum*, Bangladesh J. Agril. Res. 35(2): 217-226.
- Junqueira, A.H. and M.S. Peetz, (2012). Commercialization of *Anthuriums* in Brazil: relevant aspects of the internal and external markets. In *Anturio. Embrapa*. Castro, A.C.R., Terao D., Carvalho, A.C.P.P. and Loges, V. (eds), Brasilia. P 163.
- Lim-Ho, C. L., G. C. Lee and L. K. Phua. (1985). Clonal propagation of orchids from flower buds. Proc. 50t Asian Orchid Cong. Ed.A.N.Rao. Singapore, pp.90-110.

- Maira, O., M. Alexander and T.E. Vargas, 2009. Micropropagation and organogenesis of *Anthurium andraeanum* cv. Rubrun. Methods Mol. Biol., 589: 3-14.
- Reisch, L. (1998). Effect of media on production of *Anthurium*. Hawaii Agr. Exp. Sta. Prog. Notes No. 94.
- Sagawa, Y. and J. T. Kunisaki. 1982. Clonal propagation of orchids by tissue culture. In: Plant Tissue Cult. Ed. A. Fujiwara, Maruzen, Tokyo. Pp. 683-684.
- Somaya, K.U., P. Narayana and K. V. Jayaprasad, (1998). Micropropagation studies in *Anthurium andraeanum* Lind. Karnataka J. Agric. Sci., 11: 466-470.
- Ullah, M. H. (1995). 2nd Int. Plant Tissue Culture Conf., Dhaka, Bangladesh.