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RESEARCH ARTICLE

ROLE OF PLANT GROWTH REGULATORS (PGRS) IN *IN VITRO* SEED GERMINATION OF *ACAMPE RIGIDA* (BUCH.-HAM EX SW) P.F.HUNT

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ABSTRACT

Acampe rigida (Buch-Ham Ex. SW) P. F. Hunt an epiphytic orchid of N. E. India, has immense floricultural appeal. It is a robust species with 60-90 cm long stem bearing beautiful aromatic flowers of pale yellow colour with transverse crimson bands. Like many other orchids of this region, this species is also considered to be endangered due to large scale denundation of forest areas. So, in order to preserve it from possible extinction, attempts are made to multiply it through in vitro culture of seeds. BAP at concentration of 2.0 mg/ L. increased the number of seeds germination.

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INTRODUCTION

Orchids are one of the most fascinating and a highly evolved member of the angiosperms and Orchidaceae is the second largest family in India (Satish Kumar and Manilal, 1994). The nearest estimates range between 3,000 to 3,500 species in over 800 genera. The North East region of India perhaps the maximum wealth in the world approximately and about 800 species is known to occur in this region (Chanda, 1992). 350 species have been reported from Assam (Arora and Mukherjee, 1983). *Acampe rigida* (Buch Ham Ex. SW) P. F. Hunt is a commonly known epiphytic orchid of N. E. India having immense floricultural appeal. It is a robust species with 60-90 cm long stem bearing beautiful aromatic flowers of pale yellow with transverse crimson bands. (Hedge, 1984) Vegetatively it resembles a robust *Vanda* and under cultivation it may be grown like a *Vanda* (Teoh Eng Soon, 1980). Like many other orchids of this region, this species is also at the verge of extinction due to large scale denundation of forest areas. Each single capsule or pod consists of innumerable wind dispersed microscopic seeds lacking any functional endophytic (mycorrhizal) association required for optimum germination (Bernard, 1909; Burgeff, 1909). In nature only 0.2-0.3% of there seeds germinates (Singh, 1992). So, in order to conserve this beautiful species it from possible extinction, seeds were cultured aseptically on MS medium using *in vitro* technique. *In vitro* techniques provide better understanding of different physico-chemical requirements that might affect growth (Arditti *et al*. 1981).

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MATERIALS AND METHODS

Fresh, mature and undehisced 200 days old pods of *Acampe rigida* (Buch Ham Ex. SW) P. F.Hunt were collected from the orchidarium of the Dept. of Life Sciences, Dibrugarh University for the present study. The pods so collected were first washed thoroughly under tap water, and then with sterile DDW using 15% "Teepool". The pod was surface sterilized by immersing it in 0.1% ($HgCl_2$) solution for 5-10 minutes inside a laminar Air flow cabinet, washed 2-3 times in sterile DDW. The pod was split open vertically along the suture and the seeds were inoculated into the flasks containing Murashige and Skoog (1962) medium with different concentrations of BAP and BAP+KN. In the first case, the plant growth regulators (PGRs) were taken as 1mg/1ml of DW made up with 1000ml of MS medium. In the second case, PGRs were taken as 1mg/10ml of DW made up with 1000ml of MS medium. In both the cases seeds were inoculated in different concentrations of BAP (1.0, 1.5, 2.0, 2.5, 3.0, mg/ lt) & BAP+KN (0.5, 1.0, 1.5, 2.0, 2.5, 3.0, mg/ lt) including control and MS ½ strength. The percentage of sucrose taken as 30mg /lt, agar 8mg/ lt, pH of the medium adjusted to 5.6 before

sterilization and autoclaved at 15lbs/(inch) pressure for 10-15mins. The inoculated flasks were stored on racks in growth room under continuous conditions of temperature (22-24 degree centrigrade), humidity (65%-70%) and under 12hr/daylight (2000-3500 lux). Some of the flasks were kept under continuous light. Some more kept initially in dark, which 10 days after commencement of germination were transferred to light. The flasks were observed at regular intervals for the development and growth of green protocorms, leaves, roots etc. These were repeatedly sub-cultured at suitable intervals and photographed whenever found necessary.

RESULTS AND DISCUSSION

From the first case, seeds inoculated in MS medium where the PGRs taken as 1mg/1ml of DW, the effect of different concentrations of BAP and BAP+KN in the initiation of leaf, shoot and root can be observed from the Table 1.

Table 1. Response of different concentrations of PGRS on seeds of *A. rigida* in MS medium

PGRs (mg/l)	Seeds turned Green within (days)	PLBs formed within (Weeks)	First Leaves (weeks)	First roots (weeks)	% of seed germination on MS medium
BAP (1.0)	14	4	10	17	50
BAP (1.5)	14	4	10	16	67
BAP (2.0)	14	4	10	17	80
BAP (2.5)	28	6	12	NIL	57
BAP (3.0)	28	8	13	NIL	23
CONTROL	14	4	10	NIL	50
½STRENGTH	NIL	NIL	NIL	NIL	NIL
BAP+KN(0.5)	14	4	8	NIL	45
BAP+KN(1.0)	14	4	8	NIL	65
BAP+KN(1.5)	14	4	8	20	80
BAP+KN(2.0)	14	4	9	22	70
BAP+KN(2.5)	28	6	12	NIL	48
BAP+KN(3.0)	28	8	13	NIL	19

[PLBs- Protocorm like bodies]

Table 2: Response of different concentrations of PGRS on seeds of *A. rigida* in MS medium

PGRs (mg/l)	Seeds turned Green within (days)	PLBs formed within (Weeks)	First Leaves (weeks)	First roots (weeks)	% of seed germination on MS medium
BAP (1.0)	7	3	7	10	90
BAP (1.5)	7	3	7	10	90
BAP (2.0)	7	4	7	11	70
BAP (2.5)	10	6	8	13	45
BAP (3.0)	10	7	9	14	30
AP+KN(0.5)	7	3	7	10	75
AP+KN(1.0)	7	3	7	10	90
AP+KN(1.5)	7	3	7	10	90
AP+KN(2.0)	9	4	8	12	70
AP+KN(2.5)	10	6	8	14	45
AP+KN(3.0)	10	8	10	15	30

Thus MS medium with only BAP showed 80% germination at 2mg/lt. But at higher concentration (BAP+KN) showed lower % of germination. Again, in number, in the long run a comparative account shows that the rate and growth of plantlets and leaf size are better at BAP (2mg/lt) than BAP+KN (1.5mg/lt). Rooting, though it is better at BAP (2mg/lt) is stunted in growth and very few in (BAP+KN). These plantlets then were sub-cultured upto BAP (4mg/lt) [i.e. BAP taken from the second case- 1mg/ 10ml of DW made up with 1000ml MS medium]. Roots started to develop within 1½ months. Similarly, seeds inoculated from the second case where the effect of different concentrations of BAP and BAP+KN in the initiation of leaf, shoot and root can be seen from the Table 2. Thus, MS medium with BAP and BAP+KN showed 90% germination from 1.0-1.5 mg/lt. From 1-1.5mg/lt concentration of both BAP and BAP+KN the duration taken for growth and development of leaf size and root length was faster in comparison to other concentrations. At higher concentrations of both, germination percentage decreased progressively--duration taken for growth and development of leaf and root is slower, smaller, stunted and fewer in number. The set kept in the dark with BAP and BAP+KN at different concentrations showed germination and formed protocorms around the same time but devoid of chlorophyll. They were transferred to light after 10 days from the commencement of germination, developed green chlorophyll after 2 days but protocorms were quite elongated in comparison. So, from the mentioned two tables (Table 1 and Table 2), MS medium with BAP and BAP+KN from (1-1.5mg/lt) of both showed 90% germination in the second case. Again, the duration taken for growth and development of leaf, root length is comparatively much better in the second case than the first case. Lastly, low concentration of PGRs promotes rooting while higher concentration inhibits its formation.

From the present study MS medium with low concentration of PGRs has found to be suitable for seed germination and seedling growth. The number of roots and shoots in both the cases decreased with increased concentrations of PGRs. Rooting was completely negligible at high concentrations of PGRs in the first case. Similar obscurations have also been reported by Ueda and Torikata (1969) who observed that the number of roots and shoots decreased with increased concentration of PGRs upto 0.6(mg/l) NAA. Low concentration of PGRs promoted 90% germination from (1-1.5mg/lt) in both BAP and BAP+KN from the second case in comparison to 80% germination of BAP (2mglt) and BAP+KN (1.5mg/lg from the first case). Again from the first case, though 80% germination was observed at BAP+KN (1.5mg/l) but in the long run seedling growth decreased at BAP+KN (1.5mg/lt) in comparison to BAP (2mg/lt), may be due to the

effect of KN. Thus growth hormones both inhibit and promote seed germination in orchids depending on type of explants and concentration used (Arditti *et al*, 1981).

REFERENCES

Arditti, J., Michaud, J.D. and Olivia A.P. 1981. Seed germination of North American orchids. Native California and related species of Calypso, Epipactis, Gooodyera, Piperia, *Platanthera.Bot.Gaz.* 142: 442-453.

Arora, Y.K. and Mukherjee, A. 1983. Ornamental orchids of N.E. India, Technical Bulletin No.5, ICAR Complex NEH Region, Shillong, Meghalaya.

Bernard, N. 1909. L'evolution daus la symbiose les orchideset leur Champignons commensaux. *Ann.Sci.Bot.,* 9: 1-96.

Burgeff, H. 1909. "Die Warzelpize der Orchideen inhre kulter und ihr leben in der pflaaza" G. Fisher Verlag Jena, pp.5.

Chanda, K.L. 1992. The Orchid scenario, *J. Orchid Soc, India* 6: 1-4.

Hedge, S.H. 1984. Orchids of Arunachal Pradesh. Forest Department, Arunachal Pradesh. pp.69-70.

Mitra, G. C. 1971. Studies on seeds, shoots, tips and stem discs of an Orchid grown in aseptic culture. *Ind. J. Exp. Bio*.9: 79-85.

Murashige, T and Skoog, F. 1962. A revised medium for rapid growth and bioassay with tobacco tissue culture. *Physiol. Plant*, 15: 472-497.

Satish Kumar, C. and Manilal, K. S. 1994. A catalogue of Indian orchids. (Ed. Bishen Singh Mahendra Pal Singh). Dehra dun., 63-87.

Singh, F. 1992a. *In-vitro* propagation of seeds "state of the art". *J. Orchid Soc. India* 6: 11-14.

Ueda, H. and Torikata, H. 1969. Organogenesis in meristem culture of *Cymbidium* 11. Effect of growth substances on organogenesis in the dark. *J. Jpn. Soc. Hort. Sci.,* 38: 188-193.

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