IN VIVO GERMINATION OF ENCAPSULATED ORCHID SEEDS

ABSTRACT

The orchid seeds are naked embryos without any nutritive tissue to support them for germination *in vivo.* In nature they germinate symbiotically when they come in contact with mycorrhiza found in the mother root tufts. Orchid capsule consists of hundreds or thousands of seeds which go waste without germination. The role of mycorrhiza is the conversion of starch into simple glucose which provides nutrition for the germinating seeds. Orchid cultivars and breeders use *in vitro* technique to culture the orchid seeds with various growth hormones. An attempt was made to grow orchid seeds *in vivo* by encapsulating them with calcium alginate similar to the production of synthetic seeds. *Epidendrum radicans* seeds were encapsulated with calcium alginate. The sodium alginate gel was prepared using Vacin and Went medium supplemented with 100ml L^{-1} CW, 2mg L^{-1} NAA and 20g L^{-1} sucrose. The encapsulation was carried out aseptically and the seeds were successfully germinated in sterilized soilrite medium.

Key words : *In vivo, symbiotic, mycorrhiza, calcium alginate, Epidendrum, Soilrite.*

INTRODUCTION

The orchid capsules contain numerous seeds, which are strongly reduced structures (Beer, 1983; Burgeff, 1911, 1936). A seed weighs between 6.3 and 0.3 micrograms. They lack nutrient tissue entirely. A small germ is surrounded by a single layered seed coat which has a very loose structure. However, a few seeds will germinate on the tufts of the mother plant by symbiotic association with mycorrhiza. Though most of the seeds are found viable only few seeds are noticed to survive in nature. *In vitro* symbiotic method of seed culture was adopted by orchid breeders for many years. This method was found laborious as the mycorrhiza was found to be specific. However, Noel Bernard (1909) made the first attempt to grow these orchid seeds in controlled condition. Bernard (1909) believed that the orchids were obligate symbionts with specific nutrients necessary for the normal germination of seeds. On the other hand, Burgeff (1909) concluded that the fungus converts the starch in the embryo into glucose and increases the osmotic concentration of the cells. In the present work an attempt is made to grow the orchid seeds by encapsulating them with sodium alginate and calcium chloride solution, similar to the preparation of synthetic seeds in the nursery beds.

MATERIAL AND METHODS

5% of sodium alginate gel was prepared in Vacin and Went medium supplemented with 100ml L^{-1} coconut water, 20g L^{-1} sucrose, 2mg L^{-1} NAA. The sodium alginate was mixed to warm VW medium with constant stirring till it formed a jelly. The jelly was autoclaved at 15Pb pressure for 15min in an autoclave and cooled. 100µM of calcium chloride solution was prepared by dissolving 100mg Calcium chloride in 100ml of pre sterilized DDH_2O, in the laminar air flow. Mature fruit capsule before dehiscence were collected from *Epidendrum radicans*. They were surface sterilized with 70% ethanol and gently flamed within the LAF. The capsules were cut open with sterile scalpel and the seeds scooped out of the capsule into the sodium alginate gel. Clumps of gel with the seeds were transferred to calcium chloride solution. The encapsulated seeds were packed in polythene sachets and preserved them in the refrigerator till it was sown.

Sterilized soilrite (mixture of perlite and peat moss) was mixed with sterilized processed cocopith (Tannin content was removed by soaking cocopith in water for 48 hours). The entire mixture was treated with 2% bevestin (systemic fungicide) and filled to net pots. The encapsulated seeds were sown in net pots and transferred them to port treys. The port treys were kept in a hardening tunnel used for hardening tissue cultured plantlets, where 80-90% humidity and 25-30 ^{0}C temperature and illumination were provided.

OBSERVATIONS

The seeds responded for the condition provided and started germination within a period of 45-50 days. The seedlings obtained were allowed to grow in the same conditions. After 60 days NPK (20:20:20) solution diluted 10,000 times was sprayed daily to get better growth of the seedling. After 120 days of incubation the seedling started producing good root formation. Such seedlings were segregated and transferred to individual pots, containing broken pot pieces charcoal and moss.

RESULTS AND DISCUSSION

Orchid seeds are microscopic in structure and devoid of nutrients to germinate on its own. However, the mycorrhiza found in the mother root tufts help in converting the available starch into glucose which help in germination. Only few seeds succeed in this process and hundreds of seeds go waste without germination. Orchid breeders now a days use *in vitro* culture technique for orchid seed germination to produce new varieties. This process is time consuming and expensive. The mortality rate is very high during hardening. Since zygotic

embryos are similar to somatic embryos in the morphological features, an attempt was made to encapsulate the orchid seed, which are naked embryos, with the same technique used for synthetic seed preparation. The results obtained indicate that the orchid seeds can be successfully germinated by using this simple method, which will help in saving time and money for the breeders.

CONCLUSION

Intergeneric hybridization is possible in orchids to produce new varieties. The seeds obtained after hybridization were usually cultured in vitro this takes several years (5-6 years sometimes) for the plants to flower. This process is time consuming and laborious. present investigation will help the breeders to minimize the time of flowering, this will help in bringing out new varieties of orchids to the market at a faster rate.

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ABBREVIATIONS

VW- Vacin and Went medium

CW- Coconut Water

NAA- Naphthaline Aceyic Acid

LAF- Laminar Air Flow

NPK- Nitrogen, Phosphorous, Potassium

DDH_2O- Double Distilled Water