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Short Communication

ADVENTITIOUS PROLIFERATION OF SECONDARY AND TERTIARY PROTHALLI FROM THE PRIMARY PROTHALLI OF *PRONEPHRIUM ARTICULATUM* (HOULST. & MOORE) HOLTT.

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Abstract: In the present study the adventitious proliferation of the gametophytes of *Pronephrium articulatum* (Houlst & Moore) Holtt., has been studied. The abnormal growth was observed on the prothalli of *P. articulatum* under controlled conditions of light, temperature and humidity.

INTRODUCTION

Second only to Angiosperms, the pteridophytes, especially the ferns have proved to be an excellent experimental material for morphogenetic studies (Bir and Anand, 1982). The independent gametophytes of ferns and their morphological simplicity have enabled in to use them as experimental objects in certain areas of plant growth and development (Camloh, 1999). *In vitro* culture techniques have been used to study different aspects of germination and gametophyte development (Hickok et al., 1987]. By suitably manipulating the culture conditions *in vitro*, it has been possible to induce apogamous gametophyte and adventitious sprouts / buds (Albaum, 1938). Adventitious sprouts of a fern prothalli is an outgrowths from a single cell either inner or marginal, which develops in a similar manner to the outgrowth of a prothalli filaments from a spore. In the present study this rare and interesting adventitious sprouts formation was noted on *Pronephrium articulatum*.

MATERIAL AND METHODS

Fertile fronds of *Pronephrium articulatum* (Houlst & Moore) Holtt. Were collected from the Nilgiris hills of south Western Ghats, India. The fronds were cut into small pieces and placed on white smooth paper for 24 hrs at room temperature with fertile surface down. The liberated spores were passed through nylon mesh (40 μM) to remove the sporangial wall

material and clean spores were obtained. The spores were surface sterilized with 0.1% $HgCl_2$ (w/v) solution for 5 min. and washed with sterile distilled water for 15 min. The surface sterilized spores were inoculated on to the basal media devoid of sucrose, Knudson C (1946), Knops (1884), Mitra et al.,(1968), Moore (1903) and Murashige and Skoog's (1962). The pH of the media was adjusted to 5.8 before gelling it with 0.5 % (w / v) agar (Himedia, Mumbai). The cultures were incubated at 25°C ± 2°C under 12h photoperiod of 1200 lux density provided by cool white fluorescent tubes. Germination percentage of the spores, growth pattern and development were analysed.

RESULTS AND DISCUSSION

The spores started to germinate on 90th day and produced a green filament. The germination percentage was very low nearly 30-40 %. By day 40, the prothallus formation was initiated so that a fully developed, green irregularly lobed thalloid gametophyte appeared by the 60th day it has a small thick midrib with cushion like structure from which rhizoids emerged. Sex organs produced on the midrib region, the apical notch was absent. Sporophytes do not appear even in 3-4 months old gametophytes. The gametophytes were transferred to fresh medium, after 40 days the prothalli produce cordate green prothalli from the margin of the parent prothallus. Sharma and Sharma (1991) also observed the secondary prothalli formation, their observation was directly consonance with our result. After 30 days, tertiary prothalli generation was observed from the margins of the secondary prothalli (Fig: 1 A – D).

These were similar to the parent gametophytes of prothalli. Albaum (1938) report says that the secondary and tertiary formation was induced by the abnormal structure of prothalli (lack of apical notch). Adventitious sprouts rarely apperar on young actively growing, uninjured prothallia under optimal controlled conditions of light, temperature and humidity. They were observed only in matured and old prothalli surfaces. The very same observation observed in the present study also.

Formation of secondary generation of gametophytes from the primary prothallus on transferring to fresh medium is an interesting case regeneration through apospory. In the present study concluded that the primary prothalli marginal cells are act as spore to produce the prothalli.

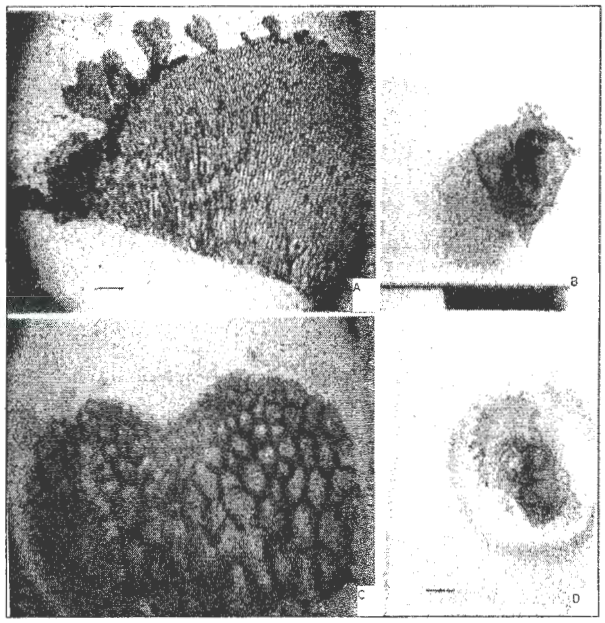

Fig: 1 Adventitious Proliferation of Secondary and Tertiary Prothalli on *Pronephrium auriculatum* (Houlst. & Moore) Holtt

A. Secondary Prothalli proliferation from the Primary Prothalli
B. Over view of Primary Prothalli and Secondary Prothalli
C. Tertiary Prothalli formation from the Secondary Prothalli – Microscopic View
D. Prothalli with Secondary and Tertiary Proliferation over view – Prothalli without apical notch

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