In Vitro Cultivation of *Gracilaria canaliculata* for Conservation and Commercial Production

Sewwandi W.H.R.M., Liyanage N.M.N., Perera S.A.C.N. and Bandaranayake P.G.C.[1*]
Department of Agricultural Biology,
Faculty of Agriculture, University of Peradeniya, Peradeniya, Sri Lanka

The current project is aimed at developing a micropropagation protocol for *Gracilaria canaliculata,* an economically important red algae for mass rearing of propagules for conservation and commercialization. Specimens were collected from Dondra Bay, Sri Lanka. Explants with meristematic growth were identified by macroscopy and microscopy; by studying cross-sections of young tips and mature stems to identify meristematic tissues. The experimental design was a CRD, with a minimum of ten replicates. Two previously recorded sterilization protocols for other seaweeds were tested with six modifications (T1-T6) under different betadine, ethanol, antibiotics, fungicide concentrations and incubation periods. Survival rates and pigment retention were recorded for 30 days. Data at ten day intervals were analyzed by a Chi-square test using SAS package. Pigment retention rate was graphically illustrated with a heatmap done using R software. A three-factor factorial experiment was designed for regeneration using previously utilized two protocols with four modifications (M1-M4) for Provasoli's enriched seawater and full MS media under different light-dark conditions, agar levels (1.5%, 0.75%, 0%) and hormones (NAA: BAP; 1:1, 2:1). Distal immature tips with meristematic growth were selected as explants. Overall, 0%, 75%, 55%, 34%, 35%, and 38% survival rates were recorded for T1 to T6, respectively and T6 (1-hr incubation in 0.5% Thiram fungicide and antibiotic mixture; 100 mgL^{-1} Streptomycin, Penicillin and 15% ethanol) was identified as the best. Explant survival rates significantly ($P \leq 0.05$) differed among sterilization methods on the 10^{th} and 20^{th} days after culturing. Explants of M4 (Full MS, 0.75% agar, light:dark; 16:8) regenerated from the meristematic regions five days after culturing. However, photosynthetic pigments of explants depleted over time resulting in a colorless but live explant in cultures. Our results revealed the possibility of micropropagation of *G. canaliculata*. Further improvements recommended for commercialized regeneration and conservation.

Keywords: Carrageenan, *G. canaliculate*, *In vitro* cultivation, Red-algae, Rhodophyta

[1]Agricultural Biotechnology Centre, Faculty of Agriculture, University of Peradeniya, Peradeniya, Sri Lanka
[*]pradeepag@agri.pdn.ac.lk