Isolation and Regeneration of Protoplasts from *Cantharellus cibarius* Fr.:Fr., an Edible Ectomycorrhizal Mushroom

Rashmi Khedkar, Anita Patil, & Mahendra Rai

Department of Biotechnology, Amravati University, Amravati, Maharashtra-444 602, India, email: mkrai123@rediffmail.com and pmkrai@hotmail.com

Mushroom mycelia are now considered as a potential source of protein, amino acids, vitamins, and minerals. Commercial production of mushroom mycelia could provide a promising source of proteinaceous food. *Cantharellus cibarius* is an ectomycorrhizal mushroom, which grows during the rainy season in association with wild bamboo in India in general and in Central India in particular.

The mushroom belongs to the order Cantharellales and the class Basidiomycotina. The mushroom is sold in open markets in the tribal pockets of Melghat (Maharashtra), Tamiya, and Patalkot in District Chhindwara; the Balaghat District of Madhya Pradesh; and the Bastar region of Chhatisgarh states of India. The ectomycorrhizal mushroom *C. cibarius* was collected from Melghat, Amravati. The gills of this mushroom were used for isolation of the fungus on "Modified-Fries-Medium." Protoplastation is interesting and useful in the case of ectomycorrhizas, in which most of the exchange of solutes takes place in apoplastic space in the hartig net area. These fungi have advantage over arbuscular mycorrhizal fungi because they can be grown on defined synthetic media and thus can be manipulated apart from the plant partner. The isolation of the protoplast can also be used to understand the mystery of symbiosis between the fungal partner and mycelia protoplast can be used to address questions pertaining to metabolic exchange.

Protoplasts of *C. cibarius* were isolated from mycelium grown in potato-dextrose broth. When mycelia were treated with a combination of macerozyme, chitinase, and cellulase and with 0.6 M sucrose as the osmotic stabilizer, the protoplast yield was 1.0×10^6 per gm fresh weight of mycelium within 3 hours of incubation. The protoplasts were regenerated in solid MFM agar plates. The regeneration frequency was between 20 and 25% and depended on culture conditions.

The main aim of isolation of the protoplast from *C. cibarius* was to study its regeneration potential.