Some Medicinal Mushrooms in Submerged Culture

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Mushrooms are promising resources of physiologically functional food and as materials for the development of medicines, pharmaceutical products such as new drugs (protein-bound polysaccharides, terpenoids, steroids, etc.), dietary supplements and healthy beverages, cosmetic products, among others. The mushroom industry is growing and new techniques are being devised to increase the number of cultivated species. Submerged pure culture techniques are used in the propagation of mycelia mushrooms in liquid media and have three main applications. It is possible to obtain in a short period of time: (1) liquid spawn for fruiting body production, (2) biomass that can be used for food and dietary supplements, and (3) biomass and/or exometabolites of medicinal mushrooms as raw materials for medicine.

In preliminary investigations the morphology and growth rate of 30 species (more than 70 strains) of edible and medicinal mushrooms from the Culture Collection of the Mycology Department of the N. G. Kholodny Institute of Botany National Academy of Sciences of Ukraine was studied on five different agar media. The optimal term of the inoculum propagation on suitable medium and temperature of incubation were determined for each culture.

The "natural" (liquid beer wort) and three synthetic liquid media with mineral and organic nitrogen sources were used for experiments. The

effect of pH medium on the mycelia growth was determined by using phosphate buffers with pH values between 3.0 and 7.5. When this factor was studied HCl and KOH solutions were used also. The inoculum has been precultured in a smaller volume of the suitable liquid media and homogenized aseptically in a Waring commercial blender for experiments carried out in shake flasks.

The underlying principle of our methodology consists of using, at each stage of inoculum preparation, mycelia in the active physiological state. Factors affecting mycelial growth of some medicinal mushrooms in submerged culture were investigated. Inoculum size, composition, and pH nutrient medium were important for the growth rate, yield biomass, and "decrease" in fermentation time.

As a result the techniques for laboratory scale submerged culture were developed for some medicinal mushrooms such as *Agrocybe aegerita* (Brig.) Sing., *Auricularia auricula-judae* (Bull.) Wettst., *Cyathus striatus* (Huds.) Hoffm., *Daedaleopsis tricolor* (Bull.) Bond. et Sing., *Flammulina velutipes* (Curt.: Fr.) P. Karst., *Hypsizygus marmoreus* (Peck) Bigel., *Lentinus edodes* (Berk.) Sing., *Lenzites betulina* (L.) Fr., *Piptoporus betulinus* (Bull.: Fr.) P. Karst., *Psilocybe cubensis* (Earle) Sing., and others.

The results of submerged culture applications as the liquid spawn for inoculation of solid substrates to produce sporophores are presented.