

Mushroom Cultures

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Fresh Mushrooms and mushroom tissue culture in tubes

Pure Culture

Pure culture is the mycelia of selected mushrooms which are grown on a given medium and is free from contamination by other micro-organisms at any stage of its preparation. The mycelia develop from the tissues or spores that have been taken aseptically from some part of mushroom. The pure culture can be sub-cultured and can be obtained from other collection or introduced from other laboratories.

Tissue Culture

The culture is prepared in an aseptic chamber such as the laminar flow using inoculating needle or scalpel which is first sterilized before used. At the beginning of the inoculation process, the pointed end of the tool is sterilized over a burner for a few seconds and let to cool for a while. One selected oyster mushroom which

is of high quality, for example, is cut into two parts and tissues from the inside part is taken and transferred onto the media.

The rationale in taking the inside part of the mushroom, instead of the outer part is because it is clean and free from contamination. In some cases, as in *Auricularia* spp., the fruiting body is soaked in alcohol before inoculation.

Single-Spore Culture

Parent mushrooms with the desired qualities are chosen and the spores collected. Spore collection can be a tiring task, but an example of a simple technique of single-spore isolation is by collecting the spores on a medium in a petri-dish. A sterilized wire-mesh is put on the petri-dish with the lid taken. A mushroom with the gills on the wire mesh is left for about 2 seconds. The culture is then incubated at a suitable temperature. After 2-3

days, the spores can be picked by an inoculating needle under a stereo microscope. This technique has been used in the isolation of *Pleurotus* spp. and *Lentinus* spp. However, with the knowledge of the reproductive system in mushrooms, not all single spore cultures can be truly fertile. At least two compatible monospore cultures are needed to produce a fertile dikaryon mycelium in which the cells are binucleate.

Multispore Culture

There are many ways of producing dikaryotic cultures. This can be done either by combining compatible monospore cultures or by germinating a mass of spores together by a process known as multispore culture. The spores are inoculated on the media and left for a few days to germinate. The hyphae fuse and many dikaryons are produced which then grow radially on the surface of the media. A distinct mycelial barrage is formed, and as a result, sectors of mycelial growth are obtained. This is a productive culture or pure line and can easily be multiplied by transferring the middle mycelia of each sector onto another medium. These productive cultures can be tested for cropping and the type that produce the desired qualities can be selected.