# Oyster Mushroom Cultivation

# Part II. Oyster Mushrooms

Chapter 7

**Cultivation Modes** 

# **BOTTLE CULTIVATION**

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# The New Substrate Container



Figure 1, 2. Plastic bottles in use for mushroom growing

Oyster mushroom cultivations on logs, on shelves and in bags are discussed earlier in this chapter. Now, the latest container used for oyster mushroom growing is a polypropylene bottle. Propylene bottles were first favored by spawn suppliers since they are heat-resistant and thus, autoclavable, endurable and thus, reusable, and easy to handle.

Mushroom growers with experience and some specialized knowledge of mushroom growing and sterile techniques can make use

of bottle culture systems for production of spawn and mushrooms. The system, however, might be impractical for growers who have just started mushroom growing or those who use pasteurized bulk substrates or composted substrates that are not appropriate for bottling. Favored are the small particle-sized growth media types such as sawdust, spent grains and grain hulls. In addition, the initial set-up cost of the system may be too high for many small-scale growers to adopt. Still, some growers may be able to develop some viable ideas from this up-to-date growing method.

# Floor Plan for the Efficient Use of Space

Commercial large-scale mushroom operations usually have two complexes, one for substrate preparation and the other for growing. An ideal floor plan for mushroom bottle preparation complex is shown in Figure 3. One can see the structure is designed to minimize the length of pathways, a design feature that minimizes contamination risks. Sterilized substrate in the bottles harbors neither harmful nor beneficial microorganisms to the mycelial growth. That means, the first comer can occupy the entire substrate in the bottle. That accounts for why sterilized substrate bottles are so vulnerable to diseases when they are exposed to air-borne contaminants before mushroom mycelia colonize them. Growers are advised to maintain the highest levels of hygiene and sanitation in mushroom operations.

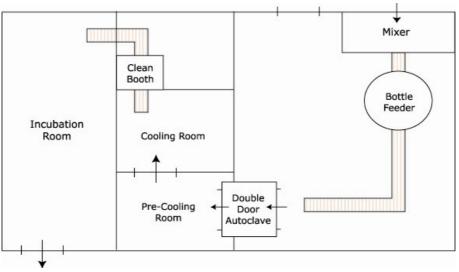


Figure 3. Floor plan for an efficient mushroom operation

# **Oyster Mushroom Cultivation in Bottles**

A bottle cultivation system employing sawdust as the growth medium would be similar to a sawdust-based spawn production system. The differences between the two include the material used as inoculum and the stage that occurs after incubation. In spawn production, inoculum is spawn master ('starter spawn') and mycelia-colonized sawdust become spawn. In bottle culture the inoculum is regular spawn and the colonized substrate is encouraged to produce fruiting bodies.

## Substrate preparation



Figure 4. Substrate mixing

The substrate materials should be in particles small enough to run smoothly into the bottle. Sawdust from hardwood or broad-leaf trees such as poplar, alder, and cottonwood are preferred. Sawdust from softwood trees like Douglas fir can be used after a three to four month-outdoor fermentation process during which the phenol compounds are dissipated. Growers are advised to use mature sawdust, but not overly aged material that may contain heat- resistant bacteria and substances unfavorable for mycelial growth.

The same substrate preparation recipes used for mushroom bag culture can be applied to substrate bottle preparation. Rice or wheat bran,

corncob or other cellulosic materials can be supplemented to promote mycelial growth. Although optimal substrate formulation varies among strains, generally four parts of basal ingredient and one part supplement are mixed and the final moisture content should be 65%. How can growers know whether the moisture content is appropriate or not? A rule of thumb among mushroom growers is one or two droplets should be released when they squeeze the mixture in the palm of their hand. Some growers add lime (calcium carbonate) to the substrate mixture to improve physical structure and lower acidity.

## Bottling

Prepared and moisture-conditioned sawdust mixture is loaded into the bottle feeder. Through the feeder, bottles are filled with the preset quantity of mixture. Once bottles are filled, compactors press the mixture in the bottle

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down to the pre-set height and hole-makers go through the compacted mixture.

Proper compaction gives the substrate high density, which means more nutrients will be available to mycelia and thus produce a higher yield. Vertical holes in the bottle permit even distribution of mushroom spawn to the bottom, which allows for fast, even colonization. Fast depletion of nutrients in the substrate, in turn, leads to an early fruiting.



Figure 5. Mixer to feeder



Figure 6. Bottle feeder



Figure 7. Feeding, compacting and hole making



Figure 8. Inoculation holes

## Sterilization



Figure 9. double-door autoclave

Filled bottles are loaded into an autoclave. Commercial scale autoclaves have double doors: one for entry and the other for exit. As seen on the floor plan, post-sterilization contamination risk from exposure to outside air is almost removed since sterilized bottles are removed from the exitonly door in the well-controlled, dust-free cooling room.

Growers are advised to make sure the autoclave has enough water and fuel so that sterilization will not be interrupted. Bottles should be sterilized at  $121^{\circ}$  or 15 psi for 60-90 minutes (from the point the temperature or the pressure reaching  $121^{\circ}$  or 15 psi). More reasonable precautions would include wearing protective gloves and removing sawdust litters from the bottle surface that could act as a contamination vector from the bottles.

# **Cooling and inoculation**

When the bottles are removed from the autoclave they should be cooled to 20 °C in the cooling room. Slow cooling is advisable as condensation occurs when hot bottles are abruptly exposed to cool air. This is why some mushroom operations have a pre-cooling room that is used before the cooling room. Bottles ready to inoculate are moved from the cooling room through a small window into the clean bench on the skate wheel conveyor. Before inoculation, the inside of the laminar flow hood must be disinfected with an ultraviolet lamp and 70% alcohol. The floor must be mopped with 10% bleach. Some large-scale farms have an air-shower before entry to the inoculation area. The highest sanitation is required since one might grow weed and disease fungi inside contaminated bottles. Visitors and workers should take off their shoes and wear clean clothing when entering this part of the operation.



Figure 10. Autoclave door to the pre-cooling room

Figure 11. Wheel conveyer from cooling room to inoculation room

Figure 12. Inoculator

## Spawn run

Inoculated bottles are hauled to an incubation room, where temperature and humidity is maintained at 17-18 °C and 65-70%, respectively. The spawn run is strain-dependant, but usually takes 20-25 days. Ventilation time and frequency vary largely depending on room temperature, humidity and the number of bottles. Growers can determine ventilation time and frequency by measuring the CO<sub>2</sub> concentration. The maximum upper limit of CO<sub>2</sub> concentration for mycelial growth is 3,000 ppm. During incubation, it is critical to perform a close examination of the bottles and look for any contamination. When unnoticed, contaminated bottles can ruin all the hard work involved in substrate preparation, inoculation, and incubation. Before fruiting, some growers opt for removing aged mycelia on the top part of the bottle.



Figure 13. spawned bottles at the incubation room



Figure 14. Good colonization

# Fruiting

When 90% of the substrate in the bottle is colonized, they are brought to a growing room or exposed to a lower

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temperature (such as when one incubates and grow mushrooms in the same place). Fruiting is induced by low temperature or high humidity as the mycelia shift into reproductive growth from vegetative growth. Growth parameters for mushroom development are the same as with mushroom bag or shelf cultivation.



Figure 15. Pinning



Figure 16. Fruiting



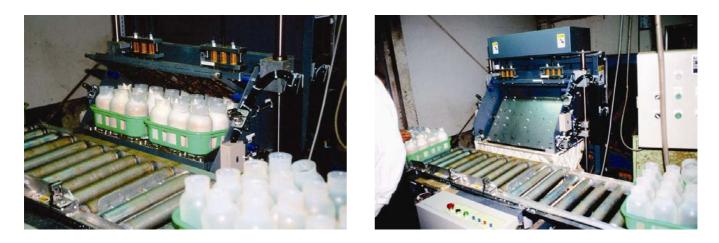
Figure 17. Fruiting bodies in the bottles



Figure 18. Fruiting bodies ready to harvast

# Emptying

After harvest, bottles are loaded into the emptying machine. This de-bottler first removes spent substrate and then washes the emptied bottle with air or water. The emptying area should be far from the growing facilities since the used mushroom substrate might harbor spores of weed or disease mold.



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#### Figure 19, 20. De-bottling

This new growing method saves much labor by automating the whole production process. Mushrooms can be "manufactured" all through the year in microclimate controlled rooms. This allows for a predictable and stable cash flow. However, as one may imagine, the initial set-up cost is too high for most beginning growers. In addition, as mushrooms are mass-produced and spawn is also self-produced in the system, skilled sterile techniques and strict hygiene practices are required. Wise growers employing different cultivation methods could use their creativity to adopt the good points of a bottle cultivation system. Growers are advised to "start small and smart but grow big."