Sawdust-bag cultivation technologies for mushroom production

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Introduction

Since its establishment a millennium ago in China, natural log culturing for mushroom production persists in many countries. Although it is a simple methodology of wood-decomposing mushroom cultivation, fruiting bodies require more than a year to form; moreover, much destruction occurs to the surrounding living forest stands. In 1970s, an innovative technique was developed in Taiwan in which a bag culture containing agricultural wastes and wood saw dusts were used in the replacement of logs. This has since brought about a revolutionary breakthrough in mushroom cultivation, the advantages of which not only enhance mushrooms particularly shiitake yield, but also to shorten cropping cycles of mushroom production. Not surprisingly, this technique quickly became pivotal to the cultivation of edible mushrooms.

To grow edible and medicinal mushrooms, sawdust derived from various trees is used. However, the kind of sawdust most suitable for growing these mushrooms depends on the individual mushroom species, such as *Lentinula edodes*, *Flammulina velutipes*, *Pleurotus* spp., *Auricularia* spp., *Ganoderma* spp., *Hypsizigus marmoreus*, *Agaricus braziliensis*, *Agrocybe cylindracea*, *Tremella fuciformis*, *Hericium erinaceus*, etc. Based on the species, fruiting conditions and substrate formulations for various mushrooms also differ. In this report, I introduce a general procedure which is useful as a benchmark for mushroom cultivation on sterilized sawdust with particular emphasis on the use of these illustrate techniques of bag culture focusing specifically for oyster mushroom and shiitake cultivation.

Procedure of bag culture for mushroom cultivation

Bag culture for mushroom cultivation requires six steps, including mixing sawdust and other nutrients/ingredients, filling bags, sterilization, inoculation (spawning), incubation (spawn run), and fruiting. The general procedure is shown as Figure 1.

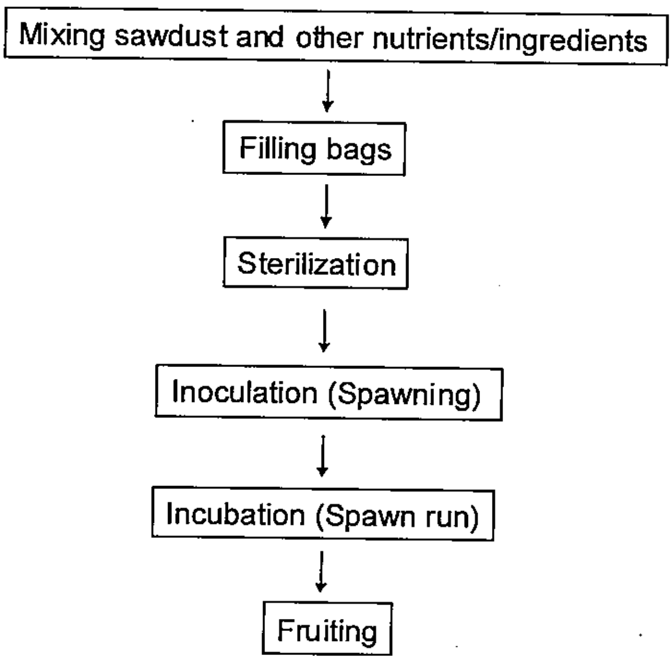

Fig 1. General procedure of mushroom cultivation on sterilized sawdust filled in plastic bags.

1. Preparation of a mixture of sawdust and other nutrients/ ingredients

In general, sawdust should be moistened then stacked for 2 to 3 days. If the fresh sawdust is not suitable for the desired mushroom growth, it should be stacked for several weeks or several months. The sawdust pile should be turned regularly to remove detrimental components such as resins and phenolics.

To speed up spawn running and increase mushroom productivity, mushroom growers add nitrogen-rich additives such as rice bran, wheat bran, soybean meal, or corn flour to the sawdust. Using calcium carbonate or ground power of burned oyster shells, the pH value of the substrate is adjusted to 6.0–6.5.

Sawdust and other substrate ingredients are mechanically stirred in a mixer equipped with an apparatus that adds water. Based on the size of the sawdust and supplements, the water content of the substrate is adjusted to 60–67%.

2. Filling the bags

A cultivation substrate containing mixtures of saw dusts and other ingredients is filled into polypropylene bags by a semi-automatic filling machine or by hand. Different countries employ different types of bag with/without filter patches depending on their filling systems. For example, a cylindrical bag (9–10 cm in diameter and 30–40 cm in height) is most frequently used to match the filling machine in Taiwan. The bag is filled with 900–1500 g substrate mixtures based on the desired mushroom species. After the bag is full, a hole is made with a stick in the middle of the substrate mixture. This hole allows spawn inoculated to the button of bags. The wider distribution of spawn enables mycelia to grow both directions (from the top and button) to the substrates, which end up faster colonization during its spawn run in the bag culture. Finally, a plastic ring is secured outside the bags, the bag top is pulled through the ring, and the mouth is plugged with a cotton ball. Heat-sealing or plastic lids are acceptable alternatives.

3. Sterilization (heat treatment)

The objective of sterilization is to diminish contaminants (mostly keep away from microorganisms, e.g. bacteria and fungi) that hinder spawn grown in the substrate. Two heat-treatment methods are used for the sterilization of the substrates: high-pressure sterilization (121℃) and normal pressure sterilization (below 100℃). In normal pressure sterilization, the temperature sterilization chamber should reach 98–100℃ in 3 hr, maintain this temperature for 4 hr using steam, and then end the steam treatment for 1 hr before pulling the bags from chamber for cooling.

During high pressure sterilization, the substrate temperature must reach 110℃ in 1.5 hr, bring the substrate temperature level to 100℃ for 30 min to remove cool air, then increase the temperature to 121℃ for 1 hr and preserve the same temperature for 1 hr with steam, and lastly end the steam treatment for 30 min and pull the bags from autoclave for cooling.

4. Inoculation (Spawning)

Spawn is the mushroom propagating material containing mushroom mycelia with a substrate mixture that promotes spawning. For the inoculation step, the sterilized substrate must first be cooled in a clean room equipped with an air shower, air filter, and mechanical cooling system. When the substrate temperature is cooled to 25–27℃, the substrate is ready for spawning. Prior to spawning, the spawn requires careful examination to ensure the substrate is free from contaminants. Depending on the desired mushroom species, the rates of spawn are 1–5% of the wet weight of the substrate.

5. Incubation (Spawn running)

For spawn running, the sealed inoculated bags are moved to an incubation room where the temperature can be adjusted according to the different stages. The spawn running time is different for each species of mushroom and also varied depending on the size of bag, the amount of spawn, strain used, and the temperature in the incubation room. The running of spawn usually takes 0.5 to 3 months for mycelia to fully spread throughout the substrate before the bags can be moved to a mushroom house or a cropping room for fruiting.

6. Fruiting

Initiating the fruit body and managing the mushroom crop varies according to mushroom species. The common strategies for inducing fruiting of mushroom mycelia include temperature fluctuations, high humidity, water soaking, ventilation, light, and physical shocks in the mushroom house or cropping room. Changes in environmental conditions are necessary to make the growth of mushroom transition from a vegetative to reproductive stage. The mushroom house for cropping is constructed differently according to the mushroom species and the mushroom growers' capital. Some mushroom species, such as *Pleurotus* spp., *Auricularia* spp., and *Agaricus* spp., can be grown efficiently and less expensively in a mushroom house without equipped with any environmental controls; whereas, some mushroom species are prone to the natural climatic changes which can greatly affects its production stability. Thus, these environmental sensitive mushroom species can only be grown in some seasons of the year. With environmental controls, however, they can be grown throughout the year.

Cultivation of oyster mushroom

Oyster mushroom, *Pleurotus* genus, grows on a wide variety of ligninocellulosic of wood wastes. According to a survey by Poppe, J. (2004), about 90 kinds of agro- or forest wastes are useful to oyster mushroom production. For small scale farms on limited budgets, oyster mushrooms are a good choice for beginning mushroom cultivation because they are easier to grow and more productive than other species.

Species selection of oyster mushroom cultivation

More than eight kinds of oyster mushrooms are cultivated in the world. Before deciding which oyster species to grow, you should consider several factors, including available agro-waste materials, appropriate facilities, costs of necessary equipment, the level of skill required to manage the life cycle of the mushroom, and the market for that species (Beetz and Greer, 1999). Some oyster mushroom species, such as *P.* citrinopileatus, *P. sajor-caju*, and *P. cystidiosus*, do not require a temperature below 20℃ for initial fruiting. Therefore, they can be grown efficiently in a mushroom house without environmental control. As for market conditions, *P. ostreatus* is one of the most popular mushrooms.

Sawdust sources

The frequently used sawdust is a mixture of different tree species from hardwoods. Without stacking, pine tree sawdust is not recommended because its resins inhibit mycelial growth. If sawdust is unavailable, rice straw/wheat straw and sugarcane bagasse serve as an alternative substrate for some *Pleurotus* species, such as *P. sajor-caju* or *P. citrinopileatus*, and produce a high biological efficiency as well.

Formula of cultivation substrate

The supplements are added to sawdust at 15% to 50% by dry weight of total substrate. Some species require lower nutrients while others require more nitrogen to produce a high mushroom yield. The additives used are mainly the by-products of agriculture, such as rich bran, wheat bran, or soybean meal.

Incubation temperature and time:

In Taiwan, most mushroom growers cannot afford expensive facilities and equipment to prepare substrates. They are not capable of producing their own spawn, either. To resolve this, several professional producers make bag substrate that has been inoculated with mushroom mycelium and sell such bags to mushroom growers. These bags are incubated at an optimum temperature of 20–25℃ in darkness since spawn running does not require light. Based on the bag size, substrate, spawn rate, strain, and temperature, full colonization takes 15–25 days.

Fruiting:

Once mycelia have fully grown through the substrates, it is ready for the formation of fruiting body – the mushrooms. In general, most oyster mushrooms do not require extra time to reach maturity. The bags with full grown mushroom mycelia may be treated two ways during the process of initiating of fruiting formation. One is to cut 4–6 slits on the sides of the bags; another is to take the cotton plug out of the mouth of the bag and strip the bag or make an opening. To establish optimal fruiting conditions, the air temperature should be maintained at 15–20℃ and the concentration of CO_2 in air is kept at the level of 1000–1500 ppm as manipulate/control by the ventilation of fresh-air. Meanwhile, the relative humidity is maintained at above 85% by the frequent addition of water in the mushroom house environment. The newly formed fruiting bodies/mushrooms can be firstly harvested within 7–10 days after the bag is cut for initiation of fruiting formation. Mushrooms are picked carefully without disturbing other developing mycelia. A continuation of 2 to 5 flushes can be harvested.

Cultivation of shiitake

Since shiitake is the third most cultivated edible mushroom in the world, I take shiitake as another good example to use sawdust-bag culture as an improved technology for mushroom production. Shiitake has earned considerable consumer demand as a fresh or dried mushroom product. It is cultivated in two ways: traditionally by natural logs and improved technology by using sawdust-bag culture. The growers can make their own decision to choose which cultivation substrate materials – pending on availability, cheapness and good recycling use of agro-wastes/leftovers for green production. Natural logs can produce high-quality shiitakes with good management, however, sawdust-bag culture has been developed and can be managed efficiently to keep high yield and with shortened fruiting cycles, thus a continuation of 3 to 6 flushes can be harvested in single cropping. This can be seen, despite a greater capital investment and more skillful management of sawdust-bag culture than natural log culture, sawdust has been evolving becoming the primary technology for shiitake production method in many countries. Figure 2 shows the pictures of the shiitake production on natural log and sawdust-bag.

Strain selection for optimal shiitake production:

Most shiitake production either cultivating in natural logs or sawdust-bag cultures is kept under natural climate without temperature control. A key factor to running a successful business for shiitake production is selecting an appropriate strain which can well adaptive to local climate. Since each strain has its proper temperature range for to initial the primodia and the duration of production period, relevant information should be acquired prior to buying its culture or spawn. According to the preferred temperature during the fruiting period, shiitakes are classified to low temperature (<10 ℃), medium temperature (10–20 ℃), high temperature (>20 ℃), and wide-range temperature (5–35 ℃) strains. In addition, the performance of shiitake strains varies in terms of substrate selectivity, growth rate, fruiting quality, and yield. (Chen 2005)

Fig. 2. Shiitake mushrooms grow on logs (left) and on sterilized sawdust bags (right).

Substrate for shiitake fruiting production:

Non-aromatic hardwoods are usually preferably choice as raw substrates for shiitake production. Rapid-decomposing hardwoods (i.e. alder, poplar, cottonwood, willow, sweetgum, and aspen) are better than slow rotting woods (i.e. oak, ironwood, mahogany). However, denser hardwoods produce more shiitake flushes/harvests than those on faster decomposing hardwoods. In general, fresh sawdust does not require go through stacking, if the sawdust contains components that would cause inhibition to mycelial growth, stacking should be conducted for up to several months.

Fresh sawdust from the trees of *Acacia confusa* Merr., *Trema orientalis, Liquidambar formosana* Hance may be used without prior fermentation. When the sawdust is moist enough, it is ready for substrate preparation.

The most commonly used substrate formulation/recipe for shiitake production is composition made of sawdust contains 15–25% rice bran (dry weight of sawdust) and 1% $CaCO_3$.

Spawn run and browning

The time for spawn running depends on the size of the sawdust bag culture, amount of spawn, strain used, and temperature. To achieve proper spawn run and browning, a period of 45 to 90 days for spawn running in the bag is necessary. After 30 to 45 days (1.5Kg/bag), mycelium shall be able to develop throughout the bag. At the end of incubation period, the entire surface of the substrate turns brown, indicating that mycelium is ready for fructification.

The browning process (oxidation of surface mycelium, also called skin formation) allows the mycelium on the bag surface to aggregate and toughen, thereby forming a layer of hyphae resistant to desiccation.

Fruiting and harvest

While the shiitake mycelium is ready for fruiting, growers take the cotton plugs out of the mouth of the bag and use a sharp knife to cut away the top of the plastic bag (partially), it should be careful not to cut too deeply and damage the mycelium. Another method is stripping the bags. The sawdust substrate block exposed to air is vulnerable to contamination under natural conditions.

Temperature fluctuations or water soaking is a common strategy to induce primordia. Soaking sawdust blocks or providing a physical shock (i.e. turning the blocks upside down) is required for subsequent flushes. Soaking allows water to rapidly displace carbon dioxide in the substrate air spaces, providing enough moisture for one flush of mushroom. Many Taiwanese growers turn the blocks upside down after the first flush as a common practice.

During harvest, shiitakes are picked by hand and the residual stalk stubs are cut off from the substrate. Some shiitakes are sold as fresh while others are dried in an oven or under the sun as dried shiitakes which can be preserved for long shelf life for storage or sale in the market.