Shiitake Cultivation

Part I Shiitake

Chapter 4

Shiitake Bag Cultivation

SHIITAKE BAG CULTIVATION

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Shiitake (*Lentinula edodes*), a mushroom of culinary delight and medicinal benefits, was traditionally cultivated on natural logs. Although shiitake spontaneous-log cultivation was discovered in China, Japanese farmers have been instrumental in developing shiitake cultivation on natural logs as a science and an industry. For nearly half a century, from 1940's until 1986, Japan was the leading producer of shiitake worldwide. As late as 1983, Japan accounted for 82.8% of the entire shiitake world production. A dramatic change in the global dynamics of shiitake production occurred in 1987. China overtook Japan, for the first time, as the number one shiitake producer in the global market by producing 178,800 tons. What most contributed to the quantum-leap in shiitake production in China was the invention of bag cultivation using sawdust as a growing substrate. In particular, the success came through the use of cylindrical sawdust bags in Gutian, Fujian. The Gutian model was quickly replicated in many parts of China with local adaptations (Fig. 1A). The well-known Biyang model today is one particular version of this system (Fig. 1B). As Daniel Royse (2002), the current leading authority on shiitake production is leaning towards sawdust bag cultivation. Bag cultivation technology makes it possible to produce shiitake all year around to meet market demands.



Figure 1. Cylindrical sawdust bags for shiitake growing A: Gutien model B: Biyang model

Types of Shiitake Cultivation Processes

Table 1 shows the cultivation processes commonly used by shiitake growers. Descriptions of other modifications can be found in Chang and Miles (2004), Chen, Arnold and Stamets (2000), and Oei (2003).

	Spawning	Spawn run	Fruiting	Bag	Growing cycle
Log cultivation	Localized	In nature (under shelter)	In nature (under shelter)	No bag	Slow
Bag cultivation					
A. Cylindrical developed in China / the substrate is usually compressed	Localized	Indoor (little environmental control)	Outdoors	Cotton or foam plugs (no air space in bag)	Moderate
B. Sawdust blocks 5-6 kg per bag / developed in the U.S. - brown in bag - brown out of bag	Through	Indoor (control)	Indoor (control)	Micro-filter breathing window / air space in bag / bags sealed	Fastest
C. Sawdust slates 15 kg per bag / developed in Europe	Localized (eg. liquid spawn injection)	Indoor	Indoor	No air space in bag / both micro-filter and foam plug are used	Fast

Table 1. Commonly used shiitake cultivation processes



Figure 2. Types of shiitake cultivation A: Natural-log cultivation (the U.S.) B: Cylindrical sawdust synthetic logs (Tibet, China) C: Sawdust-block cultivation (Fungi Perfecti, the U.S.) D: Sawdust-slate cultivation (Europe)

Preparation of Shiitake Sawdust-bag Cultivation

Strain selection

Shiitake strains are temperature-dependent. Strains are classified according to their preferred temperature during the fruiting period (Table 2). The growth rate during spawn run also differs according to the different strains, and results in either short or long durations for the mycelial maturation. Some short-duration strains take only 60 days to mature while longduration strains require 90 days. Malformation of mushrooms may occur as a consequence of farmers attempting to force fruiting too early.

Table 2. Shiitake strains classified according to fruiting temperature ($_{\rm C}$)				
Strain	China (Wu (ed), 2000)	General (Oei, 2003)	Taiwan (Chang and Miles, 2004)	
Low temperature strains	< 10	around 10	-	
Medium temperature strains	10-20	10-18	10	
High temperature strains	> 20 20 or > 20 20			
Wide-range temperature strains	5-35 (for example, China-Stamets 2)			

It is important that a strain is selected that matches a grower's specific needs. Shiitake strains vary widely not only in fruiting temperature, maturation characteristics (early or late; shorter or longer production time), but also in substrate selectivity, growth rate, fruiting quality (size, thickness, color and fragrance, etc.), yield and ecological adaptability to extreme temperature. The strains used for natural log cultivation are different from the strains that are used for sawdust bag cultivation. In sawdust bag cultivation, the strains used for the brown-in bag procedure and the strains used for the brown out of bag procedure are different. Unfortunately, some serious losses in yield have occurred because spawn makers have sold new strains that produce well on natural logs but produce very low yields when cultivated on sawdust. Some strains perform better on a substrate of corncob while others perform better on a sawdust substrate (Oei, 2003).

Substrate selection

Selection of the tree species for sawdust cultivation should be done carefully. Fresh sawdust that has not been aged can be used for production of shiitake only if it is from high quality tree species, such as those graded 4, excellent, by the FAO (Oei, 1996). Oak (*Quercus* spp.), chinkapin (*Castanopsis* spp.), hornbeam (*Carpinus* spp.), sweetgum (*Liquidambar* spp.), poplar (*Populus* spp.), alder (*Alnus* spp.), ironwood (*Ostrya virginiana*), beech (*Fagus* spp.), birch (*Betula* spp.), and willow (*Salix* spp.) are examples of commonly used non-aromatic broadleaf hardwoods. Sawdust from tree species of lower quality has to be

strates for shiftake cultivation				
	hardwood sawdust	100kg		
	wheat or rice bran	23-25kg		
Formulation A	gypsum	2.5kg		
(Wu, 1993)	calcium superphosphate	0.5kg		
	sucrose	1-1.5kg		
	water	100-140kg		
	hardwood sawdust	100lb (or 64gal)		
Formulation B	wood chips	50lb (or 32gal)		
(Stamets, 1993,	rice or rye bran	40lb (or 8gal)		
p.162)	gypsum (calcium sulfate)	5-7lb (or 1gal)		
	water	60%		
Formulation C	Monterey pine saw- dust (<i>Pinus radiate</i>)	6 part		
(The Forestry Research Institute of	hardwood sawdust (beech or poplar)	3 part		
New Zealand)	grain (barley)	1 part		

Table 3. Formulation of sawdust-based sub-

trates for shiitake cultivatio

aged before it can be used successfully (Oei, 1996; Royse, 1997; Ting, 1998; Wu *et al.*, 1995). Farmers should of course select locally available and inexpensive resources and use it accordingly. As an example, fermented *Eucalyptus* sawdust has been used successfully for shiitake production in Australia by Dr. Noel Arnold.

Substrate formulation

Table 3 shows examples of commonly used sawdust-based substrate formulations. Non-aromatic hardwoods are usually used. In addition to hardwood, utilization of pine is a subject of great interest, since pine is a readily available forestry resource. Supplemented pine-hardwood substrate (Table 3. formula C) was used successfully by the Forestry Research Institute of New Zealand to produce shiitake with satisfactory results (Stamets, 1993). Agricultural wastes, such as cottonseed hulls, corn cobs, bagasse, straw, coffee residue can also be used as alternative basal ingredients¹. The C/N ratio of the substrate must be around 25 at spawning (inoculation). For substrate formulations, growers can read Miles and Chang (1989) and Oei (1996, pp.198, 200) and Chapter 4 of this book (2005). A high water-holding capacity of the substrate combined with good aeration usually gives good results. If the substrate is too wet, the air flow in the substrate will be clogged. If the water collects at the bottom of the bag, the substrate is too wet.

¹ For more details, see ALTERNATIVE SUBSTRATE OF SHIITAKE in chapter 4.

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Substrate sterilization

The method used for substrate sterilization depends on the nature of bags (polypropylene or polyethylene²), bag size, and, the nature and amount of the substrate. For 2-3kg sawdust-based substrate in polypropylene bags, sterilization can be done in autoclave for 2 hours at 121 °C. Normal pressure sterilization is also possible, and this method is commonly employed in Thailand and China. Thai farmers generally utilize drums for sterilization and Chinese farmers use kilns³.

Shiitake Production on Sawdust Bag Cultivation

Spawning

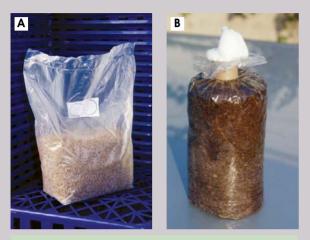


Figure 3. Various bags A: Partly filled bags B: A fully filled bag

In general, top or localized spawning, in which spawn is left on or near the surface of the substrate, is used in China, Asia, and Australia. Through spawning in which the spawn is thoroughly mixed with the entire substrate, in heat-sealed bags is used in the U.S. Larger bags with micro-porous breathing filters, partly filled with the substrates, allow the manipulation of mixing the spawn thoroughly with the substrate by mechanical shaking or manual manipulation (Fig. 3A). Smaller bags with ring necks and plugs such as those used in Asia and Australia are filled full with substrate and no air space is left in the bag. These smaller bags do not lend themselves easily to procedures for through spawning (Fig. 3B).

Caution should be taken to avoid mold contamination during spawning. Not more than 5% of the bags should be contaminated. If the contaminated molds are near the top of the bag, it is likely that the contamination gained entry during inoculation. If the contamination occurs on the bottom or side, farmers should check the bottom of the bag to see if it is sealed properly or if the

side of the bag has been punctured. If the mold is scattered throughout of the substrate, the substrate sterilization duration might have been insufficient, or the spawn used might have been contaminated (Oei, 2003). A new grower may be tempted into a money-saving scheme and may buy only a small amount of grain spawn. When he inoculates the purchased spawn into some grain he has sterilized himself to multiply the amount, unfortunately the whole lot is often contaminated, and his money saving scheme actually wastes money.

Spawn run (mycelial growth and maturation)

All shiitake strains show optimal mycelial growth during the spawn run at 25 °C. The duration of the spawn run is usually 1-4 months. Although mycelia can grow in darkness, light exposure in the first three weeks of spawn run is critical for the browning of the mycelial blocks. Some light in day/night cycles towards the end of the spawn run is conducive to the induction of primordia. It can be difficult to judge the timing for lighting during spawn run. Different approaches can be used, such as short exposure to light, e. g. 4 hours per day (Royse, 1997) or a low level of light, 50-100 lux throughout spawn run (Stamets, 1993). When the shiitake substrate is fully colonized by the white mycelia, this does not mean that the block is ready for fruiting. A mycelial maturation stage is also required. Shiitake has a complex vegetative mycelial stage composed of six stages.

- 1) **Colonization of mycelia in the substrate**: Enzymes are produced to degrade components in the complex substrate such as cellulose, hemicellulose and lignin.
- 2) Mycelia mature physiologically: Mycelia stop growing, while physiological metabolic changes occur.
- 3) **Mycelial coat formation**: Thick, white mycelial coat is formed on the substrate surface, 2-4 weeks after inoculation (spawning) in most strains (Fig. 4B). Very thick coat is formed at high CO₂ levels.
- 4) **The popcorn stage**: Clumps of mycelia are developed in some strains giving a bumpy or pop-corn like surface (Fig. 4C). Primordia are produced at the tips of some bumps (Fig. 4D). However, most bumps are aborted. Fluctuation of temperature and high CO₂ level encourages bump formation. Over-developed bumps may puncture the bag. Bumps can also be susceptible to contamination by weed molds. Some aeration should be provided when bumps are formed.
- 5) **Browning**: Two different cultivation practices are used in the American system: brown-out- of-the-bag or brown-in-bag (Fig. 4E); bags are stripped before pigmentation in brown-out of-bag (Royse, 1997), or bags are stripped after browning

² Polypropylene is heat-resistent, so suitable for high pressure sterilization. Polyethylene is divided into high pressure type and low pressure type.
³ For more details, see SHIITAKE BAG CULTIVATION IN THAILAND and SHIITAKE BAG CULTIVATION IN CHINA in chapter 4.

(brown-in-bag). Some growers remove the bag when 1/2 to 1/3 of the mycelia have become brown. The timing of bag removal is crucial, and yield can be affected if bag removal is too early or too late.

6) **Bark-forming**: In the open air, mycelia turn reddish brown at the surface and eventually forms a dark brown and dry outer protective surface which functions like a tree bark. The inner substrate becomes soft and moist as a consequence of fungal metabolic activities. Growers should be aware that a moist coat invites contamination, and should maintain 60-70% R.H. to avoid contamination.

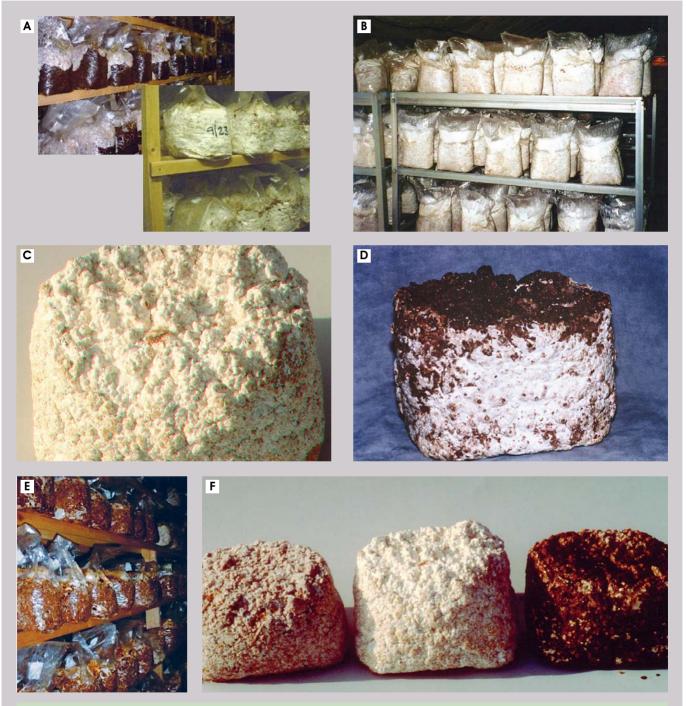


Figure 4. Spawn run process A: Bags under spawn run (Garden City Fungi) B: Fully colonized shiitake blocks (Client of Unicorn bag) C: Pop-corn like surface (Fungi Perfecti) D: Primordia formed at the tips of bumps (Fungi Perfecti) E: Brown in bag (Garden City Fungi) F: Mycelial running the popcorn stage, and browning (Garden City Fungi)

Growers should keep in mind that in shiitake cultivation, a chosen shiitake strain may prefer a certain substrate and may require a particular cultivation practice and a specific set of environmental factors for successful production. Providing an optimal environment for cultivation is vital after the best shiitake strain and substrate are chosen. Regardless of the fruiting

temperature of the strain, approximately 25° is the temperature for spawn run for all strains. Each developmental stage (spawn run, primordia initiation, fruiting) in shiitake cultivation requires a specific set of growth parameters (temperature, relative humidity, light and oxygen supply).

In the American system of heat-sealed bags with micro-filter breathing window gas exchange takes place through microporous filters. Such filters prevent rapid loss of water vapor and dehydration, thus keeping the substrate in the bags moistened. The humidity inside of the sealed bags remains high (95-100% R. H.). The design of the special micro-porous filter on the bags makes it practically care-free to manage the incubation of the spawn. It is not necessary to lower the level of accumulated CO_2 because the vegetative mycelia is tolerant of high concentrations of CO_2 up to >10,000 ppm. Ambient humidity is not critical during this period, since the bags are sealed. Growers are simply leave the inoculated substrate in the sealed bags at the natural humidity, which can be as low as 40-50% R. H.. Some growers use the same light for spawn run and fruiting, but during incubation they stack the bags next to each other to cut down the levels of available light reaching the bags (case study 1). Other growers maintain a low level of light (50-100 lux) throughout incubation (case study 2). A constant temperature of approximately 25 vis kept for all strains during the vegetative phase, although as in nature, a variable temperature also works well sometimes.

Fruiting induction for primordia initiation (Oei, 1997)

Regardless of the specific techniques used, at least some change of the environment is necessary for the transition from vegetative phase to reproductive phase (Table 4 and 5).

When the shiitake mycelia are fully matured, fruiting is induced. The following actions promote fruiting:

-temperature fluctuation

-high humidity

-water soaking (Royse, 1997: 2-4 hours at 12°C; Stamets, 1993: 24-48 hours)

-removal of CO2; supply fresh air through ventilation

-physical shocks (agitation, disturbance):

stab (with a long metal needle) and inject (with water)

turn the blocks up-side down

beating (natural logs)

 Table 4.
 The concept of environmental change in triggering primordia formation in fruiting for shiitake cultivation

To initiate primo	To initiate primordia in fruiting:			
Decrease in	1) temperature 2) CO ₂ concentration			
Increase in	 light intensity humidity: ambient humidity in the fruiting room O₂ supply: through increase of fresh air flow and decrease of CO₂ levels by ventilation 			

Note: See specifics on growth-parameter management in case studies.

 Table 5.
 How to trigger fruiting, by changing environmental growth parameters

1) Lower the temperature (according to the strain, see case studies)

2) Lower the CO_2 level, to $CO_2 < 1,000$ ppm, while

3) Increase O_2 supply by raising the frequency to 4-8 air exchange per hour

4) Increase the ambient humidity, such as to 60-80% R. H. depending on the strain and the type of cultivation practice

5) Increase light intensity, such as to 500-2,000 lux at 370-420 nm

Fruiting development

The illustrated guide below lists developmental stages in shiitake fruiting body formation in shiitake sawdust bag cultivation.

- primordia formation at the tip of the bump (blister) in the pop-corn stage (Fig. 4C)

- formation of young dark mushroom buttons (Figs. 5B, C and D)
- elongation of the stipes (stalk) as the buttons increase in size and becomes lighter in color (Figs. 5E and F)
- mushroom caps gradually unfold from in-rolled downward, while basidiospores develop in the fertile gills under the mushroom cap

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Detail description of developmental stages in shiitake fruiting can be found from further reading (Chang and Miles, 2004; Chen, 2001, 2004). During fruiting, fluctuating light is generally used. Some growers keep the temperature and humidity constant, while others simulate nature by fluctuating both the humidity and temperature, and this practice produces crops of higher quality. At the grower Fungi Perfecti, the constant and at times drastic fluctuation of humidity, during fruiting, not only improves mushroom quality but also discourages mold contamination. Several times a day, the humidity is allowed to fluctuated between 70-90% R.H.. Shiitake caps formed under such conditions have a tougher leathery outer skin which sustain a much longer shelf life (Stamets, 2000).

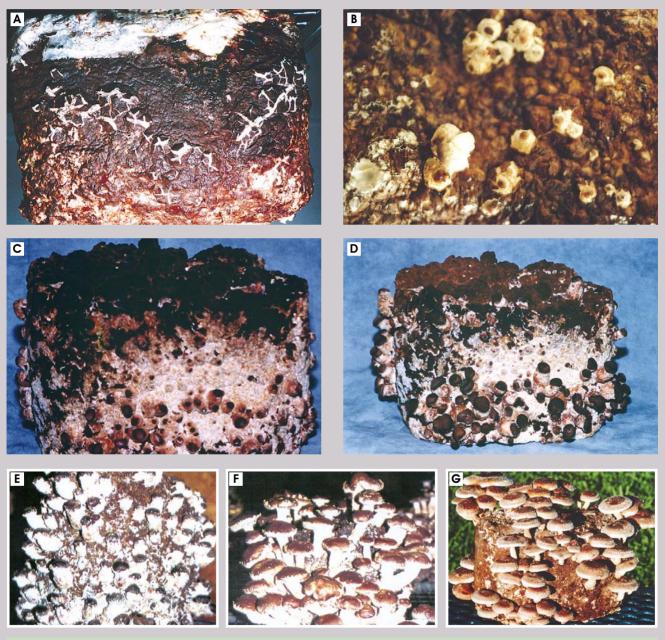


Figure 5. Fruiting development (Fungi Perfecti and others) A: Star-like cracks formed after water soaking B: Very young mushroom caps are formed C: Small young fruiting bodies D: Caps become larger, stems longer E and F: Development of fruiting bodies G: Lighter matured fruiting bodies

Harvest and subsequent flushes

To encourage better mushroom shelf life, the humidity should be lowered to 60% R.H. for 12 hours before harvesting shiitake. The desirable harvest stage for brown-in-bag technique occurs when the edge of the shiitake caps are still in-rolled downward

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(60-80% cap expansion). Shiitake are hand picked by holding the mushroom stalks and gently twisting away from the substrate blocks. To prepare shiitake for the markets, growers cut off residual stubs of stalks from the substrate and trim the ends if necessary. Remnants of residual substrate are vulnerable to mold contamination.

After harvest, the humidity is lowered to 30-50% R.H. at 21 °C for 7-10 days to induce dormancy (Stamets, 2000). The substrate blocks are then soaked for up to 12 hours to induce the second flush of the mushrooms, and up to 18 hours for the third flush (Royse, 2001). Larger bags of more substrate (5-6lb wet substrate) produce more, up to 5-6 flushes of mushrooms. Harvested mushrooms not sold as fresh shiitake are dried at 60 °C.

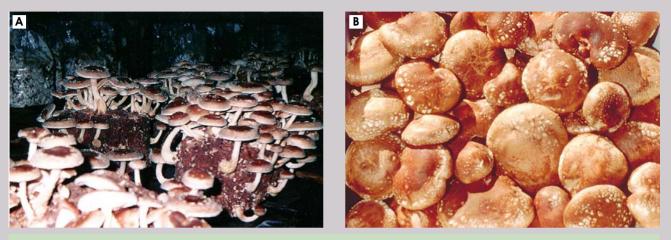


Figure 6. A: Prolific production of shiitake on sawdust blocks B: Harvested shiitake

An Overview on Sawdust-block Cultivation in the U.S.

A review of shiitake sawdust-block cultivation by Hsu, 2003 (Table 6) shows that the 250 or so serious exotic mushroom growers in the U.S. generally use two cultivation practices in growing shiitake; brown-in-bag and brown-out-of-bag. During the spawn run in the vegetative phase, the first method keeps the sawdust blocks in the bags until a brown skin (or bark) is formed before stripping the bags. In the second method, the bags are stripped much earlier when the matured mycelial blocks are still white. Curing of the stripped blocks for browning outside of the bag is done in a curing room for about 30 days. This is followed by soaking (immersion) the blocks in cold water to trigger primordia formation, the initial stage of fruiting. Fruiting bodies are formed 3-4 days later in the fruiting room. For browning in the bags, primordia formation can be initiated by spraying water or by other means such as the use of fluctuating temperatures.

The brown-in-bag procedure usually produces meatier, darker, thicker mushroom caps. The down side of this method is a longer growing cycle, so more shelf space is required. In the brown-out-of-bag system, the crop has a shorter growing cycle. Mushroom will grow faster on highly supplemented substrates that are sometimes composed of more than 45% nutrients. The produced shiitake have thinner caps and a lighter color, and special strains developed in China are used for this type of a cultivation. The different growth parameters for these two methods are shown in Table 7.

Choosing a process is a decision that depends on what kind of shiitake mushrooms consumers want in the targeted markets. A growers' skill in managing the cultivation process is another important consideration. If the grower is able to recognize physiological maturity of the white mycelial blocks, browning out of the bag is the best practice. In contrast, the maturation of the substrate in the browning-in-bag system is visually apparent, but the growing cycle is longer . Space, temperament of the grower, and hygiene management skill should all be considered. If the market already has quality shiitake that consumers are familiar with, it could be a challenge to introduce a new product. It usually takes special marketing talent and persistent effort to nurture consumers' interest for new products.

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Table 6.An overview on shiitake sawdust-block cultivation in the US, brown-in-bag vs. brown-out-of-bag (Hsu, Lou,2003, Unicorn, TX, the U.S. with author's notes*) www.unicornbags.com

	Brown-in-bag	Brown-out-of-bag
Color of the block at stripping *	Take the block out when brown	Take the block out when white
Bag used	Unicorn type 3T 14	Unicom type 14
Block weight	5-6lb	5-6lb
Substrate mix	75-80% sawdust / 20-25% supplements	55-65% sawdust / 35-45% supplements (occasionally > 45% supplements)
Type of sawdust	Oak preferred	Oak preferred
Sawdust size (typical)	30-60 mesh, 70%	30-60 mesh, 70%
Sawdust chips (typical)	5-10mm, 30%	5-10mm, 30%
Nutrients (wheat bran, rice bran, corn waste, millet, or other grain)	Locally available	Locally available
Gypsum & Calcium sulphate	0.5-2 % 0.5-2 %	
Environmental growth parameters	See table 7.	
Feature of outer brown bark	Adequately formed	Well-formed, high quality
Primordia initiation	Water spraying, etc.* (water soaking, fluctuation of temp.)	Water soaking by immersion
Type of strain	Low temperature	Fast growing (Usually of higher fruiting temp. special strains from China)
Fruiting		Mushrooms formed 3-4 days after soaking in cold water
Type of mushroom	Large cap, meaty	Thinner cap
Mushroom diameter	3 inch	2-3 inch
Number of flushes	2-3 to 4-5*	2-3 to 4-, depending on block conditions, brown bark, etc.*
Average weight of 1st flush	4-8%	8-17%
Average weight of 2 nd flush	4-8%	4-8%

Environmental Management during Sawdust-block Cultivation in the U.S.

Table 7. Environmental control of shiitake production on sawdust blocks in the U.S.; brown-in-bag vs. brown-out-of-bag (Hsu, Lou, 2003.) www.unicornbags.com

	Brown-in-bag	Brown-out-of-bag
Spawn run temperature	$20^{\circ}C$ 1 month $18^{\circ}C$ 1 month	20 °C 15-25 days Strip bag
Browning temperature	During incubation in bag	18 🕑 30 days
Primordia initiation	Strip bag, water spray, water soak- ing, fluctuation of temp.etc.	Soak by immersion in cold water
CO ₂ (during spawn run)	*no restriction before bag stripping	*no restriction before bag stripping / 8,000 ppm at curing
CO ₂ (during fruiting)	1,000 ppm and less	1,000 ppm and less
Lighting (during spawn run)	50-100 lux before bag stripping	50-100 lux
Lighting (during fruiting)	500-1,000 lux after bag stripping	500-1,000 lux

 * Notes by author: Low CO₂ level is not necessary during mycelial stage. See case study and text. No need to regulate CO₂ concentration, nor to maintain high humidity during spawn run. Easy management.

Case Study 1

Garden City Fungi, a successful grower in cultivating shiitake and other exotic mushrooms in the U.S. provided the following practical information on growing shiitake in sawdust blocks by browning in the bag (Glen Babcock, 2004).

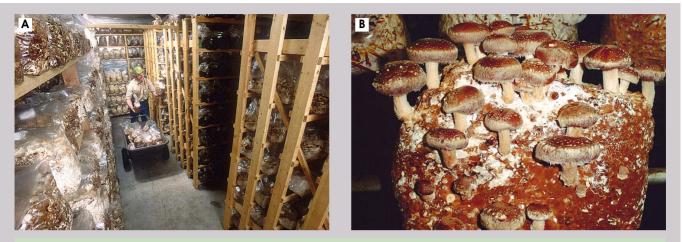


Figure 7. A: Moving shiitake blocks B: Shiitake production on sawdust-blocks

Table 8.Selection of shiitake strains and preparation of substrate blocks by Garden City Fungi, Montana, the U.S.(www.gardencityfungi.com)

Shiitake strains	Duration, 60-90 days, Garden City Fungi strains		
	60 days: GCF 1011		
	90 days: GCF 1007		
	Both work well, produce dark mushrooms with nice flecking on the cap		
Substrate block weight	6lb (wet weight)		
Bag type	Unicorn #14		
Substrate moisture content	65%		
Micro-porous filter	1 per bag		
Air space in bag	100 cubic inches $\pm = 1/20$ f the bag		
Substrate formulation	Hardwood sawdust (<i>Alder</i> or oak) 48%		
	Recycle sawdust (spent substrate) 32%		
	Millet supplement 10%		
	Bran supplement (wheat bran) 10%		
Grain spawn	v/2 cup rye-grain spawn per 6lb substrate block through spawning		
Autoclave sterilization	Bulk of 400 blocks for 6.5 hours, at 121 $_{ m C}$		

Table 9.Environmental control of growth parameters in shiitake sawdust-block cultivation, browning in bag (GlenBabcock, Garden City Fungi, 2004. www.gardencityfungi.com)

	Spawn run	Browning	Primordia initiation	Fruiting
Temperature	18-20 C constant th-	During spawn run in bag / Additional	By temp. fluctuation after	Daily fluctuation between 13-20 $_{ m C}$
	roughout day and night	browning 4-5 days out of bag when	stripping the bag / Lower	(8 hours at $13 c/8$ hours at 15-18
		necessary	temp. into 12 c at night	⁄ເ∕ 8 hours at 20 ເ_)
Light	10-12 hours per day same	Same as spawn run	10-12 hours per day	10-12 hours per day / 1,800-2,000 lux
(fluorescent light)	light source blocks (closely			by estimate blocks (spaced further
	stack-ed) receiving less light			apart) receiving more light
Humidity (R.H.)	Not important as bag is sealed	Same as spawn run	High humidity generated	85-95% all the time
	/ 40-50% R.H. in building		by humidifier	
Oxygen supply	Air exchange 0-1 / hour	Air exchange 5/hour		Air exchange 5/hour
CO ₂ level	No limit	No limit	< 1,500 ppm	< 1,500 ppm
Duration	60-90 days		4-5 days from bag strip-	2-3 flushes
	Strain-dependent over 2		ping to formation of very	6-10 days from primordia to forma-
	month		young primordia	tion of mature mushrooms
Yield				2-3lb per block on average

Notes from Garden City Fungi:

Light source: 3,000 lux or so per light, 1 bulb for every 20 ft² of incubation space. Distance of light source to block space, approximately 3-4 ft range of light. It is estimated that such lighting set-up gives 1,800-2,000 lux light intensity received by substrate blocks.

Oxygen: There is a continual flow of fresh air (frequency for air exchange 5/hour) during primordia initiation and fruiting. **Browning**: Strip the bag when browning throughout of the bag. In practice, a batch of 400 blocks are inoculated at a time at Garden City Fungi. When the time comes to open the bags, even if not all the blocks are brown throughout, all the bags are stripped. Timing is determined by blocks reach 70% or more in browning inside of the bags by prior experience. Such practice in timing is based on the fact that space is needed to start new blocks. Substrate blocks will produce mushrooms even if they are not entirely brown at stripping, as long as browning reaches 70% or more in each block. By the time blocks are done fruiting, the blocks which were not completely brown at bag stripping, will have complete browning. It takes about 2 weeks from bag stripping to finish harvest.

Primordia initiation: Allow the building to cool down to 50 °F at night, then warm up during the day. This simulates real conditions in nature where mushrooms grow in the wild. The strains used by Garden City Fungi do not form primordia well if they are not subjected to the stimulation of cool temperature in diurnal fluctuation.

Humidity: Humidity fluctuates as the temperature rises and falls. When the heater turns on in the morning, it will warm the air. As the temperature rises at this time, the humidity will fall to about 85% R.H. Once the temperature is stabilized, the humidity will return to 95%. Once again, it simulates natural conditions.

Markets for Garden City Fungi in the U.S. : Inland northwest 70% / Midwest 20% / California and other southern states 10%

Case Study 2

Fungi Perfecti (www.fungi.com), Olympia, Washington State, the U.S. (Paul Stamets, 2000), renowned worldwide. Usually brown in bag.

Humidity is high in the sealed bag during spawn run. During fruiting, fluctuate humidity several times a day (70-90% R.H.). Shiitake require lower humidity during fruiting body formation compared to some other mushrooms. Lower the humidity to 60% R.H. prior to harvest for 6-12 hours to improve shelf life.

	Spawn run (usually	Primordia initiation	Fruiting bod	y formation*	Crop cycle
	brown-in-bag)		Cold temp. strains	Warm temp. strains	
Temperature	21-27 °C	10-16′e	10-21 (c	16-27 °C	Every 2-3 weeks for 8-12 (16)weeks
Light**	50-100 lux	500-2,000 lux at 370-420nm	500-2,000 lux	at 370-420nm	
Humidity	95-100% R.H./ Inside of the sealed bag	95-100% R.H.	60-80%	% R.H.	
Oxygen through Fresh-Air exchange	0-1 per hour	4-7 per hour	4-8 pe	r hour	
CO ₂ tolerance	> 10,000 ppm	< 1,000 ppm	< 1,00	0 ppm	
Duration	35-70 days (strain dependent)	5-7 days	5-8 0	days	

*Fluctuations within the fruiting temperature ranges are beneficial to the development of the mushroom crop. **Light levels < 500 lux cause noticeable elongation of the stem.

Production of Hua-gu, the Flower Shiitake

Introduction

Prof. Yi Huang, whom I met in China in 1998, talked about his unforgettable experience of seeing hundreds of "flowers of shiitake" blooming all at once on a falling log when he was sent up the mountain for grass-root labors during the Cultural Revolution. He was so impressed by the sight, he decided right there and then to become a mushroom specialist.

Hua-gu, the flower shiitake, occurs spontaneously in nature under appropriate weather conditions during cold and dry winter months. It is not a characteristic of a particular genotype, not being a genetically inherent trait. On the contrary, hua-gu, the shiitake with an unique morphological flower-like cracking pattern on the cap, is produced artificially through manipulation of growth parameters. Hua-gu is produced at low humidity and cold temperature with fluctuation in temperature and humidity. Success in cultivation of hua-gu can bring growers considerable extra income in China. Model systems of hua-gu production can be found in Gutien county, Fujian province and Qingyuan county, Zhejiang province, in China. Hua-gu varies in quality. White hua-gu with deep and wide cracking grain and thick context (mushroom meaty part) ranks high on the grading scale, while dark tea-colored mushrooms with less pronounced cracking grain are regarded as inferior.



Figure 8. Dried flower shiitake A: High quality B: Low quality

The principle of hua-gu formation

During the formation of shiitake basidiocarps (fruiting bodies), under winter or winter-like conditions, when the young mushroom buttons reach 2-3cm in diameter, dry air and cold temperature force the pilial (cap) surface into dormancy. In such an adverse environment with drastic diurnal fluctuations of both temperature and humidity, a protective dry surface is formed on the young mushroom cap. Nonetheless, the inner portion continues to grow using water available from the substrate. When favorable growth conditions return, the surface grows at a retarded rate, while the inner portion develops at a normal pace. Under these conditions, shiitake mushroom buttons grow with alternation in dormancy and growth, and a considerable differential in the growth rate between the surface and the inner portion. In time, the rapid growth of the inner pation raptures the mushroom surface, producing a flower-like cracking pattern on the cap surface. The name, hua-gu means flower (hua) mushroom (gu) in Chinese.

The crucial factors for producing hua-gu

Low humidity, cold temperature, drastic diurnal fluctuations in temperature and humidity, diffused light, short exposure to direct mild winter sunshine, and fresh air are all conducive to hua-gu formation. Hua-gu is best produced at high altitudes and northern regions where farmers can take advantage of natural environmental conditions.

Selection of shiitake strains for hua-gu production

Growers should use low-temperature and high quality strains easily adaptable to grow at cold temperature for hua-gu production. Strains towards the lower temperature margin in the mid-temperature range can also be used.

Other examples of desirable strains for hua-gu production are: L-241-1, Jean-Yin #1, Yee-you #5, LCV141, Le 204, Le 13, 9101, 7402, 612, 9018, N-06. Strain characteristics should be thoroughly studied before cultivation. For fruiting outdoor, time of spawn-

ing should be coordinated with the maturation characteristics in order to benefit from the winter stimulation. For example, Strains L 241-4, 7402, N-06 are late maturing strains which should be inoculated early during March and April, while Yee-you #5, 9018, Le 204 are early to mid-maturing strains which should be inoculated in May-June.

Table 11. Selected strains of Lentinula edodes for flower shiitake formation (Luo, 2004)					
Strain Cultivation season Fruting temperature (°c) Duration*					
939	Autumn	8-22	120-160 days		
135 Spring 8-18 160-180 days					

* From inoculation to fruiting

Timely application of forcing hua-gu

It is important for farmers to treat young shiitake during the proper developmental stage, which is when the caps of the young mushroom buttons reach 2-3cm in diameter. If treated too early, when the mushroom buttons are smaller than 1.5cm in diameter, the fragile young mushroom buttons may die due to dehydration or freezing temperatures. If the treatments are applied too late, when the mushroom buttons have already reach 3.5cm in diameter or larger, the mushrooms do not respond readily, and only narrow and shallow cracking are formed, usually at the edge of the mushroom. The best season for applying hua-gu forcing lasts from November to the following March in Biyang, China.

Hua-gu forcing technology (Ting, 1994; Xu, 1998): Fruit in outdoor mushroom sheds. Subject young shiitake buttons of the proper developmental stage of 2-3cm in diameter to growth parameters as described (Table 12).

Table 12. Hua-gu forcing: Subject shiitake young buttons of 2-3cm in diameter to the following growth parameters.			
Dry air	65% R.H., no misting or spraying of water		
Cold temperature	8-12 (c		
Optimum	5-15°e		
Diurnal fluctuation of temperature	10 γ (in difference, desirable) Exaggerate the difference by using the covers during the day only		
Substrate moisture	55% for controlled slow growth; inject water when too dry		
Short exposure to winter sunshine	70% shading		
Proper drainage	to maintain low humidity		
Line the ground with coarse sand	already formed cracks on the mushroom cap can be re-sealed by new growth during rainy, cloudy or misty weather		



Figure 9. Flower shiitake growing from sawdust bag

Growth parameter	Spawn run	Primordia initiation	Fruiting
Relative humidity	65-70%	85%	70%
Air temperature	25 $_{ m C}$ (18-22 $_{ m C}$ if color changing)	18-22 $_{\odot}$ /8-12 $_{\odot}$ day/night fluctuation	8-18 $_{\mathrm{C}}$ fluctuation
Light	None (sunshine if color changing)	1,000-2,000 lux	1,000-2,000 lux
Air (ventilation)	1-2 fresh air exchange per day	keep fresh air	keep fresh air
Duration	60-70 days	7-8 days	20-25 days

Table 13. Specific growth parameters for hua-gu formation (Luo, 2004)

Three stages of hua-gu formation (Z. B. Yu, 1998)

Yu uses the approach in Table 14 for hua-gu forcing. Stage 1. Pre-conditioning by cold temperature to hold the growth rate back at a slow pace. Directly expose young mushroom buttons to cold air outside the bags. This is an adaptation stage to harsh environment ahead, stage 2. Hua-gu forcing parameters, stage 3. Enhancing hua-gu formation. For a case study on hua-gu formation in outdoor mushroom sheds, refer to G. H. Lo (Ting, 1998). Air temperature, humidity and mushroom cap diameter were carefully monitored and recorded during the process of hua-gu formation.

Table 14.Three stages of hua-gu forcing by Yu (1998)

Stage 1. Pre-conditioning by low temperature. Subject shiitake of the proper developmental stage (from primordia to buttons of 2cm in diameter).	
Temperature	8-12 c
Humidity	85-90% R.H. (remains high)
Stage 2. Initiating hua-gu forcing when buttons reach 2-2.5cm in diameter.	
Temperature	15 ±1 (c(8-18 (c)
Humidity	50-67% R.H. (when < 47%, apply misting of water)
Substrate moisture	50-55%
Fluctuation of temperature and humidity	
Stage 3. Enchancing hua-gu formation when caps reach 3.5cm in diameter to maturation.	
Temperature	15-25 ₍₂
Humidity	55-65% R.H.



Figure 10. Flower shiitake A: Fresh flower shiitake B and C: Dried flower shiitake

Conclusions

The current trend of shiitake production points to sawdust-bag cultivation. Growing shiitake in sterilized bags is gaining

popularity not only in Asian countries such as China, Taiwan, Singapore, the Philippines, Sri Lanka, and Thailand, but also in New Zealand, Australia, the U.S., Canada, Finland, France, the Netherlands and Germany (Oei, 2003). Growing interest is also evident in new markets in Mexico, Brazil (Renato *et al.*, 2004), Guatemala, and Peru. With the efforts of MushWorld towards encouraging shiitake cultivation as a means to alleviate poverty, it is anticipated that shiitake production will be further expanded to poverty-stricken warmer regions of the world, such as Africa and beyond.

Using shiitake bag cultivation instead of natural logs, mushrooms can be harvested faster, and the yield higher. Many types of organic wastes can be used to produce such valuable food through bioconversion. Mushrooms can be produced all year round to meet the market's demands. Shiitake bag cultivation is comparatively easier to manage and can be handled by workers, young and old, area as well as in urban districts.

It is evident that 1) shiitake varies in features and quality according to the strain, 2) different ways can be used for successful cultivation of shiitake. It is hoped that with the introduction of the practical knowledge and images presented in this review, readers will be inspired in their growing shiitake.

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