## Possibility of Shiitake Growing in Kenya

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#### ABSTRACT

Four shiitake (*Lentinula edodes*) strains were grown on different organic wastes available in Kenya. The strains used include MS-1, KV 92 and Shiitake 410 from Japan, and LE-699 from Philippines. The shiitake mushrooms were grown using the bag method.

Different substrate formulations based on locally available organic wastes were used with varying supplementation levels. The bulk substrates included Eucalyptus sawdust, sawdust from a mixture of tree species (mainly pine, cypress, eucalyptus, jacaranda, Gruvillea and flame trees), pine sawdust, sugarcane bagasse, corn cobs, water hyacinth (stems and leaves) and soybean straw.

The supplements were wheat bran and rice bran. Each of the bulk substrates was mixed with three rates of each supplement. Some formulations were also mixed with  $Ca(OH)_2$  and  $CaCO_3$ .

Half of the substrates were sterilized at 121'C using an autoclave for 2.5 hours. The other half was steamed inside a drum at 95'C for 4.5 hours. For each substrate formulation and heat treatment method, four strains were used to inoculate the substrates. All the bags were then incubated in the same growing house under room temperature (24'C). Any contaminated bags were removed from the room and records made.

After an incubation period of 34 days, slits were cut on the polythene bags. The temperature of the room was 24'C on average and the relative humidity was maintained at around 70% by spraying water on the walls and floor two times a day. The above conditions were maintained for 23 days. During this period, browning of mycelia and formation of bumps occurred. After bump formation and browning, further slitting was done. The relative humidity was raised to 95% by spraying water heavily three times a day and the temperature maintained at about 24'C.

The mushrooms were harvested after the vellum had broken and with a slightly rolled margin of the pileus. Two flushes of mushrooms were obtained.



Figure 1. A mountain of sugarcane bagasse awaiting disposal in Mumias



Figure 2. A heavy growth of water hyacinth in Nairobi dam. The water surface is fully covered

### sugar factory INTRODUCTION

Shiitake (*Lentinula edodes*) is not easy to grow in the tropics. Most substrates used in China, Japan and Korea for shiitake cultivation are not available in the tropics. The tree species that are often used, such as oaks, alders, poplar and beeches, are not common in the tropics. Cutting down of trees for log production is prohibited in many countries and if available are expensive. Production of shiitake mushrooms on logs takes a long time with relatively low yields.

The use of agricultural wastes to grow shiitake is the alternative method for many growers. The main problem with growing shiitake using the synthetic log cultivation (plastic bag) method is ensuring the right substrate formulation. The best substrate formulation will ensure high yields, high quality mushrooms and a low rate of contamination. Substrate mixtures with higher levels of supplementation (relative to the optimal supplementation level) get contaminated easily compared to those with moderate levels of supplementation.



Figure 3. Shiitake growing on Eucalyptus wood logs

Some strains do not perform well with some agricultural wastes. All the shiitake stains grown in Kenya have been imported from other countries. However, in these countries they had been used for growing shiitake on wood logs. Many growers who import the strains use them on agricultural wastes only to find that they are low yielding on these substrates. Therefore, some of the strains are not of use to farmers who do not use the wood log method of growing shiitake.

High tropical temperatures that are not suitable for the growth of shiitake and the high contamination rate in the tropics call for the proper decontamination method to lower contamination rate. This report compares the contamination risk of sterilized and pasteurized substrates on four Shiitake strains grown in Kenya.

#### MATERIALS AND METHODS

#### SUMMARY OF BAGS PREPARED:



Figure 4. A drum used for sterilizing wastes

Seven formulations with three levels of supplementation were used (Table 4) for each of four strains. For each treatment and each strain 10 replicates (bags) were made. Out of the 10 replicates, 5 were autoclaved (sterilized) and the other 5 were pasteurized in a drum.

Total No. of bags = 7 x 3 x 4 x 10 = 840 bags. Number of bags autoclaved =  $840 \times 5/10 = 420$  bags Number of bags pasteurized =  $840 \times 5/10 = 420$  bags.

The cooled substrates were then spawned with 4% spawn (wet weight basis) of the specific strain of shiitake. Spawning was done in a clean room that had been sprayed with 70% ethanol. Each bag was labeled with details on substrate formulation, decontamination method and the strain used.

After an incubation period of 34 days, slits were cut on the polythene bags. The temperature of the room was 24'C on an average and the relative humidity was maintained at around 70% by spraying

water on the walls and floor two times a day. The above conditions were maintained for 23 days. During this period, browning of mycelia and formation of bumps occurred. After bump formation and browning, further slitting was done. The relative humidity was raised to 95% by spraying water heavily three times a day. The temperature was around 24'C.

The mushrooms were harvested after the vellum had broken and with a slightly rolled margin of the pileus. Two flushes of mushrooms were obtained. The mushrooms were weighed and records made.

The research was conducted on-site at five Shiitake mushroom farms. Observations and data collection with regard to the cultivation methods, heat treatment methods and contamination rates were made on a regular basis. Additionally, questionnaires were presented to the farmers and the results compiled.

#### **RESULTS AND DISCUSSION**

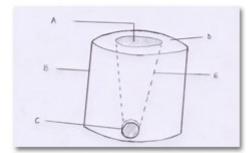


Figure 5. Drum and sawdust stove



Figure 6. Bags contaminated by green and black mould

AOpening	at	t t	he	top	of	stove.	
BMetallic		barrel		e.g	а		drum.
CMetallic	tube	making	а	horizontal	passage	for	air
DTop				of			drum.

E.....Vertical air passage formed after packing sawdust and removing empty bottle.

The cost of equipment for the various heat treatment methods i.e. sterilization using an autoclave, drum pasteurization using firewood and sawdust were determined based on best available data. Shiitake farmers gave the costs of the equipment they had bought and also the cost of treating the substrates. Numbers of contaminated bags in the farm test were recorded and the results given in relation to the substrate formulation and heat treatment method used. Time needed for full colonization of substrate was also noted from the farms. A comparison of the yields from the various strains and substrate formulations were collected.

The results given here are from the farm test carried out and include the first and second flush obtained. The substrates produced third and fourth flushes but are not included in the results as they were not produced in the time frame of the research competition. The research was initiated between 3rd of September and 19 th September 2004.

Table 1: The duration and cost of decontamination, contamination rate and colonization time using various methods of decontaminating substrates.

Method of	Sterilization	Pasteurization	Pasteurization
decontamination	using an	using a Drum &	using Drum and

	Autoclave	Firewood	sawdust stove
Duration (hrs)	7.5	4.5	4.5
Cost ( Kenya Shillings)	Autoclave (39 litres) =80,000 Power = 160 Total = 80,160	Drum = 640 Firewood = 160 Total = 800	Drum = 640 Stove = 500 Fuel = 4 Total = 1,144
Contamination rate	15%	8%	8%
Time needed for colonization	35 days	28 days	28 days

\* Duration is for decontamination of 30 bags.

The total costs given in the table above include the cost of heat treatment equipment (autoclave, drum, stove and fuel).

One batch in the drum method pasteurizes three times the number of bags that can be sterilized in one batch using an autoclave (Table 1). The drum is larger and thus can hold a larger number of bags to the advantage of the farmer. The cost of buying the autoclave is very high compared to that of the drum. The cost of electricity and firewood is equally high while the cost of pasteurizing using sawdust is 40 times lower. This method could be used to lower the production cost of shiitake thus giving a higher profit margin to the farmers.

The results show that the risk of contamination from sterilized wastes is double that of pasteurized wastes. Sterilizing wastes leaves a biological vacuum in the substrates, and thus, if a single spore of contaminant gets access to the sterilized substrate, it will multiply very fast uninhibited. Contamination, in part, was due to the lack of proper spawning equipment. This increases the risk of contaminating substrates during spawning. The rate of colonization was higher in the pasteurized wastes than in the sterilized wastes. The sterilized wastes needed about 7 days more to be fully colonized.

Formulation*	Sterilizati	on by Autoclave	Pasteurization by drum		
	Production Cost	No. of contaminated bags	Production Cost	No. of contaminated bags	
A1	460	1	410	0	
A2	510	3	460	1	
A3	430	1	380	0	
B1	440	1	390	0	
B2	490	2	440	0	
В3	425	0	375	0	

# Table 2: The cost of production of 10 bags and rate of contamination of substrate treated by sterilization or pasteurization.

C1	520	1	470	1
C2	530	2	480	1
C3	510	1	460	0
D1	380	2	330	1
D2	430	3	380	1
D3	350	2	300	0
E1	500	1	450	1
E2	560	4	510	2
E3	460	2	410	0
F1	500	2	450	1
F2	560	4	510	2
F3	460	2	410	0
G1	410	2	360	0
G2	440	2	390	1
G3	400	0	350	0

\* Formulations are listed in Table 4.

\* Production cost of 10 bags in Kenyan shilings.

\* Number of bags contaminated our of 10.

The production costs of sterilized substrates were found to be higher than that of pasteurized substrates. The production cost of the substrate with high supplementation level was high because the cost of bran as a supplement is high. The substrate formulation with the least amount of bran had low production cost. Some bulk substrates, e.g. sugarcane bagasse, bean straw, water hyacinth (Eichhornia crassipes) and corn cobs, are free. Rice bran and wheat bran are expensive. The substrates that were available for free were better than those that were required to be purchased or were transported from a far site. Other additives like urea,  $CaCO_3$  and  $Ca(OH)_2$  also have implications on the production cost. The cost of pasteurizing using sawdust as a fuel source is cheaper than any other fuel source.

 Table 3. Yields and Biological Efficiencies Obtained from growing different Shiitake strains

 on the different substrate formulations.

Formulation*		S-1 10 bags)		-699 10 bags)		AKE 410 10 bags)		/ 92 10 bags)
	Yield (kg)	B.E(%)	Yield (kg)	B.E(%)	Yield (kg)	B.E(%)	Yield (kg)	B.E(%)
A1	2.96	37	2.88	36	2.61	32.625	2.23	27.875
A2	2.78	34.8	2.64	33	2.38	29.75	2.49	31.125
A3	2.35	29.35	2.22	27.75	2.12	26.5	2.01	25.125
B1	2.32	29.05	2.18	27.25	1.96	24.5	1.96	24.5
B2	2.79	34.9	2.18	27.25	1.78	22.25	1.78	22.25
В3	2.58	32.25	1.82	22.75	1.66	20.75	1.71	21.375
C1	3.20	40	2.98	37.25	2.61	32.625	2.40	30
C2	3.20	40	2.68	33.5	2.69	33.625	2.03	25.375
C3	2.84	35.5	2.60	32.5	2.35	29.375	2.22	27.75
D1	3.68	52.55	3.12	39	2.45	30.625	2.31	28.875
D2	3.74	53.4	3.22	40.25	2.51	31.375	2.02	25.25
D3	3.13	44.7	2.88	36	2.40	30	1.76	22
E1	2.72	34	2.04	25.5	2.25	28.125	1.88	23.5
E2	2.79	34.9	2.12	26.5	2.01	25.125	1.96	24.5
E3	2.41	30.1	1.72	21.5	1.88	23.5	1.72	21.5
F1	3.03	37.85	2.48	31	2.26	28.25	2.36	29.5
F2	2.84	35.55	2.36	29.5	2.10	26.25	2.23	27.875
F3	2.54	32	2.30	28.75	1.86	23.25	2.43	30.375
G1	3.20	46.1	2.36	29.5	2.01	25.125	1.93	24.125
G2	3.25	46.4	2.13	26.625	1.78	22.25	1.80	22.5
G3	3.03	43.25	1.84	23	1.63	20.375	1.65	20.625

\* Formulations are listed in Table 4

The yields shown in these results were from the first two flushes obtained. The first flush was

found to account for the highest yield with the second flush being smaller. This is attributed to the availability of nutrients in the substrate. During the first flush, the mycelia has stored enough food prior to fruiting. The fruiting bodies from the first flush were observed to be bigger than those produced from the second flush. This is also due to the amount of food stored by the mycelia. For the drum method, the first flush accounts for 54.4% of the total yield obtained from the two flushes.



Figure 7. Shiitake grown from sawdust, rice bran and water hyacinth



Figure 8. Shiitake grown from sawdust, rice bran and water hyacinth



Figures 9 and 10. Shiitake in the 2nd flush

There was no substantial difference between the yields obtained from the drum and the autoclave methods. When using MS-1, the total yield from the autoclave method when using MS-1 was 30.56kg while for the drum method the total yield was 30.82kg.

Strain MS-1 was most suited for cultivation of Shiitake in Kenya using the bag technique. In the two flushes when grown on sugarcane bagasse it produced a biological efficiency of 54%. If the substrate had been allowed to produce 2 or 3 more flushes, the BE from the total yield from all flushes would be expected to average around 80%. Strain MS-1 seems to be most adapted to growing on the substrates and at the high temperature.

The strain LE-699 was the second best substrate and it performed well on sawdust and sugarcane bagasse based substrates. Strain KV-92 grows well on logs (as reported by one grower) but produced poor quality and many premature fruiting bodies on our particulate materials.. Shiitake 410 produced moderate yields on the bags but was reported to produce low yields on the logs by growers.

Contamination in the tropics is a serious problem to mushroom farmers. The temperatures during the day may reach 28'C which makes growing shiitake a problem without air conditioning. Even fruiting during the hot seasons is almost impossible. The best method of decontamination was pasteurization at a temperature just below 100'C. Pasteurization using a drum is the most appropriate for poor farmers because it is cheaper and most applicable to most farmers. Pasteurization contamination rate was lower compared to sterilization. The drum was also able to

hold a larger number of bags compared to the autoclave. The drum is cheaper compared to the autoclave, which requires close to KSh 80,000. The drum costs only about 1,000KSh. To lower contamination rate the incubation room should be kept as cool as possible by spraying water several times a day during the hot days. Spraying water lowers the temperature due to the evaporation so contaminants do not grow fast.

A higher rate of supplementation leads to an increase in the rate of contamination. Considering that supplements are expensive, higher supplementation can increase the cost of production. High supplementation should be left to growers in the colder regions.

It is hoped that many new farmers will find the growing methods described in this report useful to their cultivation and that older experienced farmers would also benefit by reducing the cost of growing shiitake.

	Basal material	Supplement + Additive
A	1 78% Eucalyptus	20% Wheat Bran + 2% $CaCo_3$
1 1	sawdust	
A	2 68% Eucalyptus	30% Wheat Bran + 2% CaCo <sub>3</sub>
	sawdust 88% Eucalyptus	
A	3 sawdust	40% Wheat Bran + 2% CaCo <sub>3</sub>
в	1 78% Sawdust (mix)	20% Corn Waste + 2% CaCo <sub>3</sub>
В	2 68% Sawdust (mix)	30% Corn Waste + 2% CaCo <sub>3</sub>
В	3 88% Sawdust (mix)	10% Corn Waste + 2% CaCo <sub>3</sub>
		4%Wheat
C	1 93% sawdust (mix)	$bran+1\%Ca(OH)_{2}+1\%(NH_{4})_{2}SO_{4}+1\%$ Urea
~		10%Wheat
C	2 87% sawdust (mix)	bran+1%Ca(OH) $_2$ 1%(NH $_4$ ) $_2$ SO $_4$ +1% Urea
		1%Wheat bran+1%
C	3 96% sawdust (mix)	$Ca(OH)_2 + 1\%(NH_4)_2 SO_4 + 1\%Urea$
D	1 750/ 9 1	20%Wheat bran+4%molasses or
D	1 75% Sugarcane bagasse	sucrose+1%CaCo <sub>3</sub>
р	0 (50/ 9	30%Wheat bran+4%molasses or
D	2 65% Sugarcane bagasse	sucrose+1%CaCo <sub>3</sub>
П	2 950/ Sugaraana hagagga	10%Wheat bran+4%molasses or
D	3 85% Sugarcane bagasse	sucrose+1%CaCo <sub>3</sub>
E	1 Pine Sawdust (5 parts)	Rice bran (3 parts) + Water hyacinth (2
	i i ne suwaust (s purts)	parts)
E	2 Pine Sawdust (4 parts)	Rice bran (4 parts) + Water hyacinth (2 $parts$ )
		parts) Rice bran (2 parts) + Water hyacinth (2
E	3 Pine Sawdust (6 parts)	parts)
F	1 Sawdust (mix) (5 parts)	Rice bran (3 parts) + Bean Straw (3 parts)
F	2 Sawdust (mix) (4 parts)	Rice bran (4 parts) + Bean Straw (3 parts)
F	3 Sawdust (mix) (6 parts)	Rice bran (2 parts) + Bean Straw (3 parts)
G	× 1 /	Sawdust (1 part) + Wheat bran (1 part)
G	2 Corn cobs (3 parts)	Sawdust (1 part) + Wheat bran (2 parts)

G3 Corn cobs (4.5 parts) Sawdust (1 part) + Wheat bran (0.5 parts) More figures...



Figure 11. A shed constructed using locally available materials



Figure 13. Shelves made of timber



Figure 15. Mushroom house made of earthen bricks, cement walls and iron sheet roof.



Figure 17. Shiitake spawn



Figure 12. Roof insulated with cartons

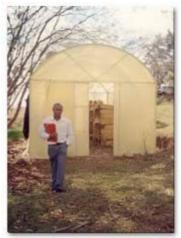


Figure 14. A green house used for shiitake growing under trees



Figure 16. Canvas protecting mushroom shed from direct light



Figure 18. Inoculation box



Figure 19. Small pins in PP bag



Figure 20. Shiitake ready to harvest