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Evaluation of cotton waste, paper waste and jatropha cake for culture of *Pleurotus sajor-caju* under different pasteurization methods

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Abstract

Identification of suitable substrate material and correct substrate pasteurization method is critical for successful oyster mushroom (*Pleurotus sajor-caju*) culture. An experiment was undertaken in a mushroom growing house to evaluate the effect of two pasteurization methods (boiling and use of Na hypochlorite) for *P. sajor-caju* culture on different substrates (cotton waste, waste paper and jatropha cake). A 3×2 factorial laid out in a completely randomized design (CRD) with three replications was used. Spawning was done at a rate of 8% and only the first three flushes were considered for the evaluation of substrate productivity. Mean number of basidiocarps (MNB), mean basidiocarp weight (MBW), biological efficiency (BE) and number of days to first fruiting (DFF) were used to evaluate substrate productivity. No fruiting was observed on jatropha cake under either pasteurization technique. Fruiting occurred on cotton waste and waste paper under both pasteurization methods. The results showed significant interaction effect ($p<0.05$) of substrate type and pasteurization method on MBW, BE and DFF. Cotton waste pasteurized by boiling method had highest productivity in terms of MNB, MBW and BE while waste paper pasteurized by Na hypochlorite solution had least productivity. However, waste paper pasteurized by boiling method took fewer DFF while cotton waste pasteurized by Na hypochlorite took the longest DFF hence delayed fruiting. Cotton waste proved to be the best substrate for *P. sajor-caju* production while boiling was the most effective pasteurization method. Jatropha cake is not suitable for oyster mushroom culture.

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Introduction

In the past years interest in mushrooms greatly increased in Zimbabwe, mainly due to the current shortage of mushrooms on the local market, which has caused prices to escalate (Mabveni, 2004) hence mushroom cultivation has subsequently become a highly profitable activity. Oyster mushrooms (*Pleurotus sajor-caju* and *P. ostreatus*) were selected by smallholder farmers because of the adaptability of their cultivation technology (Wood, 1985), and also their similarity to local indigenous mushrooms; 'Nzeve' and 'Huvhe' (*Termitomycetes*). Culture of oyster mushrooms is becoming popular throughout the world because of their abilities to grow at a wide range of temperatures and to utilize various lignocelluloses (Baysal *et al*., 2003; Khanna and Garcha, 1981). Cultivation of oyster mushrooms by the smallholder farmers would also help reduce the incidence of mushroom poisoning by providing known, well-identified edible species (Alexoupoulous *et al*., 1979).

The mushroom cultivating techniques pave the way for the future food of human beings (Ponmurugan *et al*., 2007). Growing mushrooms gives so much satisfaction and produces so much food and income that further use of this practice can result in a great complete contentment of families and villages, of which *Pleurotus* can make use of the largest variety of waste substrates with its fast mycelial growth and its multilateral enzyme system that can biodegrade nearly all types of available wastes (Poppe, 2004). For the Zimbabwean smallholder farmers and resource-disadvantaged communities, mushroom cultivation enables them to have a balanced diet at a relatively inexpensive cost (Mswaka, *et al*., 2001). Edible mushrooms rank above all vegetables and legumes (except soybean) in protein content and have significant levels of Vitamin B and C, and are low in fat (Stamets, 1993).

Mushroom cultivation also enables farmers to utilize organic substrates that would otherwise be regarded as waste products (Wood, 1985; Labuschagne, *et al*., 2000). Identification of suitable substrate material is critical for successful mushroom cultivation (Zandrazil and Kurtzman, 1982; Shah *et al*., 2004). Vast quantities of organic wastes, particularly lignocellulosic materials, are generated annually through the activities of agricultural, forest and food processing industries (Buswell, 1991). Since oyster mushroom is a first decomposer, it assists in the recycling of agro-waste, like cotton lint waste, which would potentially pollute the environment. Mushroom production is therefore a favourable income-generating project for developing countries such as Zimbabwe, from both an environmental and cost point of view (Fanadzo *et al*., 2010).

Waste paper is a threat to environmental pollution especially at academic institutions such as schools, colleges and universities. Cotton waste from ginneries exacerbates the situation, and so does jatropha cake from the biodiesel industrial sites after the processing of *Jatropha curcas* seed. There is thus need to utilize and evaluate the productivity potential of these waste materials for oyster mushroom production since they are rich in lignocelluloses, hence helping reduce environmental pollution. Oyster mushroom cultivation is highly labour intensive, short duration crop and land saving, welcomed by the poor farmers (Shah *et al*., 2004).

Successful cultivation of mushroom often requires pasteurization of the substrate, prior to inoculation with spawn (Chang and Hayes, 1978). In Pakistan, Ali *et al*. (2007) reported the use of different methods of substrate pasteurization by oyster mushroom growers which results in large variation in their mushroom production. Chitamba *et al*. (2012) suggested a different pasteurization method for jatropha cake since boiling resulted in the substrate's decay. There is therefore need to subject the substrate to different pasteurization methods so as to study its performance and conclude its ability and potential for use in oyster mushroom culture.

The present research was undertaken with the main objective of finding out the most appropriate

pasteurization method for the substrates cotton lint waste, waste paper and jatropha cake which will be helpful for further improvement in yield of *P. sajor-caju*.

Materials and methods

The experiment was carried out during the winter of 2009 in a mushroom growing house (MGH) in Chiwaridzo, Bindura, Zimbabwe. The area falls under Natural Region IIb of Zimbabwe's Agro-ecological Zones. A 3×2 factorial laid out in a completely randomized design (CRD) with three replications was used. Substrate type had three levels; cotton lint waste, waste paper (mixture of office papers and newsprints) and jatropha cake. Pasteurization method had two levels; hot water treatment with boiling water for 1 hour, and chemical pasteurization with Na hypochlorite at a rate of 20 ml per 10 litres water for 20 minutes. Cotton lint waste was obtained from Cottco Bindura while waste papers were collected from Bindura University of Science Education.

Each substrate had a total dry mass of 6 kg so that each bag would consist of 1 kg dry substrate. The substrates were put differently in metal containers filled with water and were soaked overnight so that they would achieve a moisture content of about 70-80%. They were then pasteurized by the different pasteurization methods as outlined above. The pasteurized substrates were collected using a heat sterilized fork, placed on a Na hypochlorite disinfected plastic sheet and were allowed to cool to a temperature of about 25-37°C. The wet pasteurized substrates were each divided into three equal parts making the three replications and were packed in clear Na hypochlorite disinfected plastic bags. Spawning was done in three layers at a rate of 8%. The bags were hung in the MGH and holes were punched on the bottom of the bags to drain off excess water. The bags were then left for spawn running (incubation).

Daily watering of the MGH walls and floors was done so as to maintain relative humidity during the spawn running period. Sand was put on the floor to aid moisture conservation and the room was always kept closed. A footbath with a disinfectant solution was put at the door so as to avoid introducing contaminants. The MGH was kept light free during the spawn running period. When the substrate appeared completely white as a result of successful spawn run, holes were punched on the surface of the bags to allow the pin-heads to come out. High humidity was maintained by daily watering and spraying of the bags to avoid withering of the emerging mushroom pin-heads.

The first three flushes were considered and each bag was harvested separately and the mass of the basidiocarps was recorded in grams using a digital scale. Mean number of basidiocarps (MNB), corresponding to number of basidiocarps per bag; Biological efficiency (BE), calculated as:

$$(\text{Mushroom Fresh Weight}/\text{Substrate Dry Weight}) \times 100;$$

and mean basidioarp weight (MBW) were determined for evaluation of substrate productivity. Number of days to first fruiting (DFF) was also recorded among the treatments.

The collected data was subjected to Analysis of Variance using GenSat Discovery Edition 3 package and their means were separated by Least Significance Difference (LSD 0.05).

Results

Substrate type and pasteurization method had significant effect on MBW and BE. Substrate type alone had highly significant effect ($p<0.001$) on MNB, MBW, BE and DFF. Cotton waste pasteurized by boiling water had the highest MBW and BE ($p<0.05$) but it was however not significantly different from cotton waste pasteurized by Na hypochlorite (Table 1), while waste paper performed equally lower under either pasteurization method with lower MBW and BE as compared to cotton waste. The jatropha cake substrate pasteurized by either method did not produce any fruiting bodies hence had least productivity. There was however no significant difference among the treatments and

there was no interaction between substrate type and pasteurization method on MNB ($p>0.05$), as shown in Table 1. There was significant difference ($p<0.05$) among the treatments in the number of days taken to first fruiting (DFF). However, cotton waste pasteurized by Na hypochlorite took the longest time to fruiting, followed by cotton waste pasteurized by boiling while waste paper pasteurized by boiling had the shortest time, in terms of DFF (Table 1).

Table 1. Substrate and pasteurization effects on mean basidiocarp weight (MBW), mean number of basidiocarps (MNB), biological efficiency (BE) and days to first fruiting (DFF).

Substrate	MNB	MBW (g)	BE (%)	DFF (days)
Cotton waste + boiling	208.7[a]	897[a]	89.7[a]	37.00[b]
Cotton waste + Na hypochlorite	190.7[a]	707[b]	70.7[b]	39.67[a]
Waste paper + boiling	152.0[b]	550[c]	55.0[c]	34.67[c]
Waste paper + Na hypochlorite	147.0[b]	498[c]	49.8[c]	36.33[b]
Jatropha cake + boiling	0[c]	0[d]	0[d]	*NF
Jatropha cake + Na hypochlorite	0[c]	0[d]	0[d]	*NF
Grand mean	116.4	442	44.2	24.61
F prob.: Substrate type	<0.001	<0.001	<0.001	<0.001
Pasteurisation method	0.225	0.004	0.004	<0.001
Interaction	0.471	0.013	0.013	0.021
l.s.d	22.59	84.7	8.47	1.258
CV%	10.9	10.8	10.8	2.9

Means followed by the same letter are not significantly different at p = 0.05.

*NF: no fruiting occurred on the substrate hence undefined DFF.

Discussion

Cotton waste substrate had higher productivity than waste paper in terms of MNB, MBW and BE. This study showed that the composition of substrate has a great influence on MBW and BE. Variable ranges of MBW and BE have been reported when different lignocellulosic materials were used as substrates for cultivation of oyster mushrooms (Liang *et al*., 2009). This can be attributed to the fact that cotton waste contains widely variable amounts of total N ranging from 0.25-1.45% (Chang-Ho *et al*., 1979) which promotes protein synthesis and mushroom growth, and also to high cellulose content of 95% (Collop, 2008) thus a high capacity to accumulate dry matter. Studies conducted by Tan (1981) also revealed that cotton waste was the best substrate for the cultivation of *Pleurotus spp*. The results of this study also concur with those by Adebayo *et al*., (2009) where the highest yield of *P. pulmonarius* was obtained from cotton waste. The emergence of mushrooms from the bags is purely based on the amount of cellulose present in the substrates (Sivaprakasam and Kandasamy, 1981) hence giving rise to different numbers of basidiocarps in different treatments.

Pasteurization by boiling in hot water produced the best results in terms of MNB, MBW and BE, as compared to pasteurization by use of Na hypochlorite. These findings are more or less similar to those of Ali *et al*., (2007) in which higher productivity of *Pleurotus spp*. was found in cotton waste pasteurized by steam and lower in cotton waste pasteurized chemically by formalin. Jatropha cake did not produce any fruiting bodies under either pasteurization method. These results concur with the findings of Chitamba *et al*. (2012) where jatropha cake did not produce fruiting bodies under the boiling pasteurization method.

There was significant interaction effect of substrate type and pasteurization method on the DFF. However, hot water pasteurized substrates took fewer DFF than chemically pasteurized substrates by Na hypochlorite. This can be attributed to the effectiveness of chemical pasteurization (through use of Na hypochlorite) in suppressing growth of competitor microorganisms and partly suppressing mushroom mycelia formation. Our findings on DFF of cotton waste (37) are consistent with those by Iqbal *et al.* (2005) where 37.7 was recorded for *P. sajor-caju* under the boiling pasteurization method.

Conclusion

Jatropha cake cannot be used as a substrate for *P. sajor-caju* production under either pasteurization method as evidenced by neither mycelia nor fruiting bodies formation from the present study. For optimum yields, cotton waste is a competent substrate for *P. sajor-caju* production through the use of the boiling pasteurization method. Waste paper is an equally good substrate for *P. sajor-caju* where cotton waste is scarce.

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