Comparative Study on Growth and Yield of Oyster Mushroom (*Pleurotus ostreatus*) on Different Substrates

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ABSTRACT

Agriculture in the country is transforming from subsistence to commercial farming system. Most of the time crop residues are left unutilized in the field after harvest. Burning crop debris after harvest is an age old tradition followed by farmers here in Bhutan. Mushroom growers inoculate Oyster mushroom only in rice straw unlike in other countries. Therefore, to diversify Oyster mushroom inoculation to enhance production in the country, and to find best alternative substrates other than rice straw, a comparative study on growth and yield of Oyster mushroom (Pleurotus ostreatus) on different substrates was conducted. Pleurotus ostreatus strain 'PBN' was selected species in the study. A Completely Randomize Design (CRD) with five treatments and five replications including control (Paddy straw) was used. Data were analyzed through one-way ANOVA with SPSS version 22. The treatment mean of five replications were compared using Tukey's Multiple Range Test. The yield and mycelium colonization rate differed among substrates under same climatic condition. There is significant difference in yield between control and T_1, T_3 &T_4, and no significant difference in yield between T_0 &T_2 ($P<0.01$). However, higher yield is obtained from T_2. Significant differences in colonization rate were observed among T_0, T_4 & T_3 and in pin head formation among T_0, T_1 & T_3. Yield obtained from each flush from each treatment is directly proportional to the number of pin head formed in each treatment. A total of four effective flushes from five replications were evaluated for yield analysis.

Keywords: *Oyster mushroom, Mycelium colonization, Yield, Primordia*

1. Introduction

Oyster mushroom is a saprophytic fungus. It can grow on all types of agricultural wastes throughout the year in any agro-climatic zone proper cropping shed is provided. It generates food by decomposing complex organic matter into simple compound (Chang & Miles, 1991). Unlike button mushroom, Oyster mushroom can grow directly in a substrate without composting (Atkins, 2014; Statmets, 1983). It was first cultivated in Germany as a survival mechanism during World War II (Eger, Eden & Wissig, 1976).

Oyster mushroom adapts well in dark environment (Kong, 2014), and has bittersweet smell of benzaldehyde (Beltran-Garcia, Estarron-Espinosa & Ogura, 1997). Lingo-cellulosic substrates

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are good source of growing Oyster mushroom (Sánchez, 2010). *Pleurotus spp*. are rich in medicinal and nutritional properties. Medicinally it has anti-cancerous, anti-inflammatory, antiviral, antibiotic, anti-diabetic and anti-modulator effects (Lavi et al., 2010). Nutritionally it is rich in protein, minerals and vitamin B, C & D (Panjikkaran & Mathew, 2013), and hence the popular reference to mushrooms as poor man's protein.

Pleurotus ostreatus belongs to family *Pleurotacea* and class *Agaricomycetes*. Locally it is called '*Naaky Shamung*' in west and '*Yuelay Bamung*' in eastern part of Bhutan. Like Shiitake mushroom, Oyster mushroom cultivation in the country is also expanding. At present, Oyster mushroom is commonly grown on rice straw as substrate. Oyster mushroom cultivation on rice straw is limited in its scope for expansion since farmers use rice straw as fodder for cattle, and there is also limited rice growing areas, particularly in the east.

The main objectives of this study is to explore Oyster mushroom inoculation in other agricultural wastes (crop residue) beside rice straw and to find out best alternative substrates in order to enhanced Oyster mushroom production in the country. The study also complements finding a solution to better utilization of agricultural waste.

1. Materials and Methods

The experiment was conducted from 1st week of April, 2018 to 2nd week of June, 2018 in Agriculture Research and Development Sub-Centre, Khangma. The five treatments are as follows (replicated 5 times).

T_0: Paddy straw (control)
T_1: Dried banana leaves
T_2: Dried quinoa stubble
T_3: Dried mustard straw
T_4: Dried lemon grass

2.1. Preparation of Spawn

Pure culture was developed in Potato Dextrose Agar (PDA) media (39g PDA in 1 liter distilled water) from fresh fruit body. Later it was transferred to autoclaved wheat grain and developed mother spawn. Inoculated bottles were incubated for a month in incubation room at room temperature 23°Cand 60% relative humidity.

2.2. Preparation of substrate

Different substrates were chopped at 2-5 cm length. Weigh of 10kg dry weight of each substrate were measured and filled in jute sack.

2.3. Soaking

Filled jute sacks were immersed fully under water for 15 minutes. Excess water was drained out for 5 minutes after soaking. Moisture content of each substrate was calculated manually based on

Rahman et al. (2012). Different substrates' moisture absorption capacity are tabulated below (Table 1)

$$MC\ (\%) = \frac{Wet\ wt\ of substrate - Dry\ wt\ of substrate}{Wet\ wt\ of substrate} x100$$

Table 1.Moisture absorption capacity of different treatments

Treatments	Dry weight (kg)	Wet weight (kg)	Weight gain (kg)	MC (%)
T_0	10	27	17	63
T_1	10	28	18	64
T_2	10	26.3	16.3	62
T_3	10	26.8	16.8	63
T_4	10	26	16	62

2.4. Sterilization

Steam treatment was provided to all treatments in steel barrel for 2 hours after ballooning of plastic sheet covering the mouth of barrel. It was monitored and measured with TR-71wf thermo-recorder, and cooled for 30 minutes by spreading on clean plastic sheet before inoculation.

2.5. Inoculation/spawning

Samples of size 2 kg were prepared in clean polypropylene bag from each substrate. Layer method of spawning was followed with spawn rate of 100g per 2 kg substrate. Three holes on each incubated bags were prepared for air circulation.

2.6. Incubation

Inoculated polypropylene bags were transferred to incubation room. Room was treated with 70% ethanol with 3 days interval. Room temperature and relative humidity inside room was maintained at 20-25°c and 65% respectively.

2.7. Experimental design, data collection and statistical analysis

The experiment was laid out in Completely Randomized Design (CRD). Starting date of mycelium growth, days required to complete substrate colonization, days of pin head formation and yield were recorded in MS Excel sheet. Yield was measured with digital physical balance. Recorded data were analyzed by one way ANOVA and treatment means were compared through Tukey test (P<0.01) in SPSS version 22.

3. Result and Discussion

Yield, mycelium growth colonization rate and pin head formation were analyzed. The total yield for analysis was obtained from four effective flushes. Significant difference in yield were observed amongst control and T_1, T_3 & T_4 whereas, there is no significant difference in yield between T_0 &T_2 (P>0.01). However, higher yield was obtained from T_2 compared with T_0 (Table 2), and the lowest yield from T_4. On an average 2kg substrate produced 484g of fresh mushroom, which is in contrast to findings by Jiskani (1999) where he reported that 1 kg fresh mushroom can be generated from 1 kg dry weight of substrate. Unlike Tan (1981) who found each bag to give two flushes, all treatments in this study gave four effective flushes successfully. Highest yield was obtained from second and third flushes compared to first and fourth. However, yield obtained from the substrate may differ with size of substrate: higher the substrate size higher the yield obtained.

Except for T_4, mycelium colonization in all treatments started from 3rd day after inoculation. Colonization in T_4 started only from the fourth day. Significant differences (P<0.01) were observed among T_0, T_4 & T_3 (Table 2). There is no significant between T_0 & T_2, yet, faster mycelium colonization rate was observed in T_0 while healthy mycelium was observed in T_2 (Table 2). Mycelium colonization inside substrate was completed within a span of 21 days after inoculation, as similarly reported in other studies (Mondal, Rehana, Noman & Adhikary, 2010; Tirkey, Simon & Lal, 2017). Rate of mycelium colonization in different substrates may be due to difference in chemical compositions and C: N ratio of the substrate as presented by Bhatti, Mir and Siddiq (1987). Myceliums survive well at room temperatures of 20-25^{o}C and room relative humidity of 60-65% (Gisleröd, 1987; Karacanci, 1997). High humidity inside cropping shed will create favorable environment for green mold (*Penicillium sp.* and *Aspergillus sp.)* that can inhibit mycelium colonization and hence, should be monitored closely.

Significant differences (P<0.001) in the duration of pin head (primordia) formation is observed amongst T_0, T_1 & T_3 (Table 2). Early pin head was formed in T_3 (24 days) and longer duration was observed in T_1 (27 days). Nevertheless, prior to one week after full colonization, primordium was formed in all inoculated bags. Similar result was stated in studies by Ahmed (1988) and Stamets (1983). Number of primordia found in each bags in each flush has direct effect on yield. Higher the number of primordial, higher the yield.

Table 2.Effect of different substrates on yield, mycelium growth, colonization rate and pin head formation

Treatments	Yield (g/bag)	Initial mycelium growth (days)	Fully colonization (days)	Pin head formation (days)
Banana leaves	448[a]	4[a]	20[c]	27[c]
Lemon grass	449[a]	3[b]	19[b]	26[b]
Mustard straw	457[a]	3[b]	17[a]	24[a]
Quinoa straw	533[b]	3[b]	18[b]	26[b]
Paddy straw	530[b]	3[b]	21[c]	25[b]
SE	7.42	0.13	0.63	0.48

[1]Values are means of five replicates. Different letters indicate significant differences between the treatments at $P<0.01$, [2] SE- Standard Error.

4. Conclusion

Oyster mushroom (*Pleurotus ostreatus*) has short cropping duration in that inoculation to harvest cycle can be completed in a month. It can grow in any agro-ecological zone if proper cropping shed is provided. However, success of mushroom farm solely depends on management practices and sanitation maintained inside cropping shed during cropping period. Mycelium colonization rate, primordia formation duration and yield varied among the treatments. Significant differences in these parameters were observed amongst the treatments.

Number of primordia formed in inoculated bags directly affected the yield. Regular watering inside cropping shed is equally essential during primordial formation period so that proper fruit bodies are formed. Though effective production can be generated from first flush to fourth flush, mushroom growers can still harvest few fruit bodies until the colonized substrate get exhausted. For *Pleurotus ostreatus,* optimum room humidity and temperature is 60-65% & 20-25^{0}C respectively during incubation period and 80-90% room humidity should be maintained during fruiting period.

Mushroom growers can utilize agricultural wastes for growing Oyster mushroom in order to use crop debris effectively and economically, and can be an ideal recommendation for utilizing farm waste as well as generate fast cash return.

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