**Short communication**

**Evaluation of lignocellulosic substrates on productivity of Pearl oyster mushroom (*Pleurotus ostreatus*)**

**Abstract**

Among the oyster mushrooms, Pearl oyster mushroom (*Pleurotus ostreatus*) is one of the important edible mushrooms cultivated in India. It can be grown on different lignocellulosic substrates. However, its productivity depends upon the nature and the composition of the substrates. Therefore, ten different agri-wastes were evaluated for their ability to produce mushroom. The trial was conducted in Centre of Tropical Mushroom Research and Training (CTMRT), OUAT, Bhubaneswar. Data on days to spawn run, pin head initiation, first harvest, biological efficiency (%), average number of fruit bodies per bag, yield (g / bag) along with morphometrics were recorded and analysed with Randomised Block Design. Maximum production of mushroom with 1366.67g/2kg dry substrate was realised from banana pseudostem followed by paddy straw. It was also observed that a range of 40 to 60 C:N ratio of the substrates favoured higher productivity of mushroom.

**Keywords: Pearl oyster, *Pleurotus ostreatus,* substrates, C:N ratio**

**Introduction**

*Pleurotus* spp are commonly known as oyster mushroom because of their oyster-like structure. These are the second most popular mushroom in world after button mushroom. These are known as wood fungus and in India it is popularly known as “Dhingri”. These mushrooms are grown in around 25 countries, with China, South Korea, Japan, Italy, and Taiwan being the primary producers. It is also commercially grown in the states like Chhattisgarh, Odisha, Punjab, Maharashtra, Tamil Nadu, Bihar, Gujarat and North Eastern states like Meghalaya, Tripura, Mizoram and Assam. Among the 38 species cultivated worldwide, Pearl oyster mushroom (*Pleurotus ostreatus*) is one of the important cultivated mushrooms. It belongs to family Pleurotaceae of Phylum Basidiomycota. It grows naturally in the temperate and tropical forests on dead, decaying wooden logs. It is a common edible mushroom cultivated using straw and other substrates. Different agricultural and forest by products including straw of paddy, wheat, ragi; stalk and leaves of maize, jowar, bajra, cotton, sugarcane; peanut shells; dried grasses; paper; coffee waste and synthetic compost of button mushroom with rich cellulose, hemicelluloses and lignin contents are used for cultivation of oyster mushroom in large scale. However, yield of oyster mushroom largely depends upon the nutrition and nature of the substrates. *Pleurotus ostreatus* is a potential source of protein along with vitamins and minerals. It has also got different pharmaceutical properties such as antitumor, antioxidant, anti-platelet aggregating, antimicrobial and antiviral activities. Keeping the above importance facts, efforts have been undertaken to assess the nature of substrates and their effects on the productivity of this mushroom.

**Materials and methods**

**Oyster mushroom bag preparation**

All the experiments were conducted in the growing rooms of the Centre of Tropical Mushroom Research and Training (CTMRT) in the Department of Plant Pathology, Odisha University of Agriculture and Technology (OUAT), Bhubaneswar. The test fungus (*Pleurotus ostreatus*) was procured from ICAR- Directorate of Mushroom Research, Solan (HP). The fungal culture was multiplied in Potato Dextrose Agar (PDA) medium. To evaluate the efficacy of substrates, ten different lignocellulosic substrates were collected from the locality. The dried substrates were chopped to a size of 2 inch with help of chaff cutter followed by soaking for six hours. The substrates were maintained 65% moisture for preparation of oyster mushroom bags. The bags were filled with 2kg substrates on dry weight basis in polyethene bags. About 3% of 15days old spawn were applied in layer method on the substrate. The prepared bags were incubated on different tiers of the cropping room with appropriate temperature, humidity and light for mycelia development.

The cumulative yield of each replication was recorded which was represented as weight (g) per unit of dry substrate. The yield was expressed in terms of biological efficiency (B.E.) and calculated using following formula:

$$Biological efficiency \left(\%\right)=\frac{Fresh weight of mushroom (g)}{Dry weight of substrate (g)}×100$$

**Biochemical analysis of substrates**

**Estimation of carbon percentage**

The carbon content of the substrates was determined according to report of Nelson and Sommers (1982). The percentage organic carbon content was then calculated according to the formula below;

|  |  |
| --- | --- |
| C (%) = | M x (Vbl - Vs) x 0.003 x 1.33 x 100 |
| G |

Where,

M =Molarity of FeSO4

Vbl = ml FeSO4 of blank titration

Vs = ml FeSO4 of substrates titration

g= mass of substrates taken in gram

0.003= milli-equivalent weight of C in grams (12/4000)

1.33 = correction factor used to convert the wet combustion C value to the true C value since the wet combustion method is about 75 % efficient in estimating C value , (i.e. 100/75 = 1.33)

**Estimation of nitrogen**

Similarly, the nitrogen content of the samples was determined according the procedure described by Bremner and Mulvaney (1982).

**Cellulose determination**

The amount of cellulose was determined using procedure followed by Philip and Adrian (1998).

**Lignin determination**

Klason method was used for determination of lignin content of substrates.

 **Statistical analysis**

The field trials were analysed using randomized block design (RBD). Mean value of each character were worked out by dividing the total value with number of observations.

**Results and Discussion**

The analysed data in table 1 revealed that days to spawn run in the substrate varied from 17.93 days to 28.53 days. Minimum days requirement for spawn run was 17.93 days observed in sugarcane bagasse followed by paddy straw (21.47 days) and finger millet stalk (21.53 days). Moreover, sesame stalk (23.40 days), maize stalk (23.93 days), rice husk (24.00 days) and banana pseudo stem (24.07 days) were statistically at par with each other. The maximum time 28.53 days was taken by saw dust. Early pinhead initiation was observed in finger millet stalk (25.93 days) followed by paddy straw (27.40 days), sugarcane bagasse (28.33 days) and maize stalk (28.33 days) respectively. Days to first harvest of mushroom was recorded from finger millet stalk (32.60 days) which is statistically at par with that of sugarcane bagasse (32.67 days), paddy straw (33.80 days) and sesame stalk (33.80 days). The range of oyster mushroom yield varied from 1366.67 g to 383.33 g from 2 kg dry substrate. Highest yield of mushroom recorded 1366.67 g from banana pseudo stem with 246.40 number of mushrooms. The second highest yield of mushroom 1246.67 g observed from paddy straw with 240.27 numbers of mushrooms which was statistically at par with finger millet stalk (1203.33 g, 248.87 mushrooms). Minimum yield 383.33 g harvested from paper with lowest number of mushroom (69.00). Similar trend was observed in biological efficiency with a range of 19.17 percent to 68.33 percent. It was interesting that maximum fruit body weight of mushrooms 5.70 g with lower biological efficiency (43.83 %) recorded in case of saw dust as compared to the other substrates.

**Table 1 :Evaluation of ligno-cellulosic substrates for production of oyster mushroom (*Pleurotus ostreatus*)**

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| **Sl. No.** | **ligno-cellulosic substrates**  | **Days to spawn run** | **Days to pin head initiation** | **Days to 1st harvest**  | **BE (%)** | **Average numbers of fruit bodies/ bag** | **Yield (g)/ 2 kg dry substrate** |
| 1 | Paddy straw | 21.47 | 27.40 | 33.80 | 62.33 | 240.27 | 1246.67 |
| 2 | Sugarcane bagasse | 17.93 | 28.33 | 32.67 | 53.33 | 218.33 | 1066.67 |
| 3 | Maize stalk | 23.93 | 28.33 | 34.47 | 49.67 | 197.27 | 993.33 |
| 4 | Paper | 25.27 | 31.60 | 41.13 | 19.17 | 69.00 | 383.33 |
| 5 | Sesame stalk | 23.40 | 29.73 | 33.80 | 55.83 | 240.93 | 1116.67 |
| 6 | Groundnut haulm | 24.73 | 32.00 | 36.07 | 38.17 | 184.73 | 763.33 |
| 7 | Rice husk | 24.00 | 33.13 | 39.73 | 42.50 | 174.93 | 850.00 |
| 8 | Banana pseudo stem | 24.07 | 30.67 | 35.20 | 68.33 | 246.40 | 1366.67 |
| 9 | Saw dust | 28.53 | 36.93 | 43.20 | 43.83 | 153.93 | 876.67 |
| 10 | Finger millet stalk | 21.53 | 25.93 | 32.60 | 60.17 | 248.87 | 1203.33 |
|  | SE(m)+ | 0.23 | 0.34 | 0.49 | 1.62 | 2.03 | 32.36 |
|  | CD(0.05) | 0.67 | 1.02 | 1.45 | 4.81 | 6.04 | 96.14 |

The data on morphological characters of mushroom harvested from different substrates are presented in table 2. The morphology of mushroom has a great role in yield of mushroom per bag. Pileus size of mushrooms varied from 55.00 mm to 72.67 mm. Maximum pileus size (72.67 mm) was observed in mushrooms harvested from saw dust which is at par with that of banana pseudo stem (71.53 mm). Minimum pileus size (55.00 mm) observed in case of finger millet stalk and sesame stalk which were at par with that of rice husk (55.40 mm) and groundnut haulm (58.67 mm). Similarly, maximum stipe length was observed in case of saw dust (31.73 mm) followed by paper (29.67 mm). However, paddy straw (22.47 mm), banana pseudo stem (22.53 mm) and rice husk (23.07 mm) were statistically at par with each other with respect to stipe length. Stipe diameter of mushrooms harvested from different substrates varied from 5.67 mm to 7.80 mm. Maximum stipe diameter (7.80 mm) recorded in the mushroom harvested from banana pseudo stem was statistically at par with that of paddy straw (7.13 mm), groundnut haulm (7.13 mm), finger millet stalk (7.07 mm) and saw dust (7.00 mm).

**Table 2: Morphometrics of oyster mushroom (*Pleurotus ostreatus*)as influenced by ligno-cellulosic substrates**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Sl. No.** | **ligno-cellulosic substrates**  | **Pileus size (mm)** | **Stipe length (mm)** | **Stipe diameter (mm)** |
| 1 | Paddy straw | 61.07 | 22.47 | 7.13 |
| 2 | Sugarcane bagasse | 64.53 | 27.47 | 6.80 |
| 3 | Maize stalk | 63.07 | 28.67 | 5.67 |
| 4 | Paper | 67.60 | 29.67 | 6.00 |
| 5 | Sesame stalk | 55.00 | 24.07 | 6.53 |
| 6 | Groundnut haulm | 58.67 | 18.53 | 7.13 |
| 7 | Rice husk | 55.40 | 23.07 | 6.47 |
| 8 | Banana pseudo stem | 71.53 | 22.53 | 7.80 |
| 9 | Saw dust | 72.67 | 31.73 | 7.00 |
| 10 | Finger millet stalk | 55.00 | 23.80 | 7.07 |
|  | SE(m)+ | 1.32 | 0.50 | 0.32 |
|  | CD(0.05) | 3.91 | 1.49 | 0.96 |

Data in the table 3 revealed that maximum and minimum percent of carbon was observed in rice husk (67.22 %) and paper (42.51%) respectively. It was observed, that all except paper, possess more than 50 percent carbon. Nitrogen percentage among the substrates varied from 0.40 % to 2.02 %. The nitrogen content of seven substrates out of ten recorded less than one per cent except groundnut haulm (2.02 %), banana pseudo stem (1.41 %) and paddy straw (1.19 %). Similarly, carbon nitrogen ratio (C:N ratio) of the substrates varied from 28.05 to 133.18. The C:N ratio of saw dust, rice husk and paper recorded more than 100. The cellulose content of the substrates varied from 29.20 % (rice husk) to 89.14 % (paper). Similarly, lignin content varied from 8.46 % to 31.37 %. However, maximum and minimum cellulose lignin ratio was recorded from paper (10.54 %) and maize stalk (1.25 %), respectively.

**Table 3: Chemical composition of ligno-cellulosic substrates**

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| Sl. No. | Ligno-cellulosic substrates | Carbon (%) | Nitrogen (%) | Carbon nitrogen ratio | Cellulose (%) | Lignin (%) | Cellulose: Lignin |
| 1 | Paddy straw | 63.6 | 1.19 | 53.45 | 32.61 | 14.52 | 2.25 |
| 2 | Sugarcane bagasse | 56.34 | 0.74 | 76.14 | 41.30 | 22.41 | 1.84 |
| 3 | Maize stalk | 62.63 | 0.84 | 74.56 | 39.20 | 31.37 | 1.25 |
| 4 | Paper | 42.51 | 0.40 | 106.28 | 89.14 | 8.46 | 10.54 |
| 5 | Sesame stalk | 64.34 | 0.84 | 76.60 | 43.20 | 12.18 | 3.55 |
| 6 | Groundnut haulm | 56.66 | 2.02 | 28.05 | 32.60 | 25.65 | 1.27 |
| 7 | Rice husk | 67.22 | 0.56 | 120.04 | 29.20 | 17.52 | 1.67 |
| 8 | Banana pseudo stem | 59.35 | 1.41 | 42.09 | 53.32 | 13.75 | 3.88 |
| 9 | Saw dust | 53.27 | 0.40 | 133.18 | 38.60 | 27.3 | 1.41 |
| 10 | Finger millet stalk | 57.23 | 0.92 | 62.21 | 37.30 | 13.83 | 2.70 |

Oyster mushrooms have the ability to grow on a wide range of agricultural wastes with variable yield potential. Hence, ten ligno-cellulosic wastes of different crops evaluated to ascertain the variability with respect to mycelia growth as well as productivity. All the experiments were conducted in the growing room of CTMRT, OUAT, Bhubaneswar. On the basis of days to spawn run in different substrates evaluated varied from 17.93 days (sugarcane bagasse) to 28.53 days (saw dust). As compared with other workers, spawn run in different substrates varied 13 to 20 days (Patra and Pani, 1995; Ahmed, 1998 and Jiskani, 1999). Quicker spawn run in sugarcane bagasse than paddy straw reported by Iqbal et al., 2005 and Mosisa et al., 2015 corroborated the present result. Apart from mycelia growth in substrates, there was wide variation of yield potential among the substrates. Realization of higher yield of mushroom from banana pseudo stem supported the findings of Iqbal et al., 2005. However, the yield potential of sugarcane bagasse as compared with paddy straw did not agree with the result of Khalaphallah et al., 2020. Low yield of mushroom from saw dust and paper may be due to the lack of porosity that allows the mycelium for respiration and ease access in the substrate in order to get nutrition (Osunde et al., 2019). Shah et al., 2004 recorded maximum biological efficiency of oyster mushroom (*P. ostreatus*) from saw dust in comparison with wheat straw but Shauket, 2012 reported highest biological efficiency from wheat straw followed by paddy straw. Higher cellulose and lignin ratio enhanced the yield of mushroom suggested by Badu et al., 2011 supported the present findings. The variation in yield of oyster mushrooms on different substrates were because of physical and chemical characteristics such as pH, water holding capacity, porosity, carbon nitrogen ratio, cellulose lignin ratio etc. The present result of higher C:N ratio of sugarcane bagasse facilitated quicker mycelia growth than paddy straw. Similarly, the present result with respect to C:N ratio and morphology in case of banana pseudo stem and paddy straw agreed with that lower C:N ratio favours the growth of mushroom as reported by Yang, 2000.

**Conclusion**

From the present experiments it was observed that a range of 40-60 carbon nitrogen ratio is preferred by *P. ostreatus* for mushroom production. Similarly, higher cellulose and lignin ratio of the substrates was preferred by the fungus to produce mushroom. Moreover, paddy straw is the preferred substrate for mushroom production as well as easy management of the wastes.

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