Influence of substrate pasteurization, bag dimensions and organic supplement practices on yield and yield attributing parameters of Indian oyster mushroom, *Pleurotus pulmonarius*

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ABSTRACT

Pleurotus mushroom, generally referred to as 'Oyster mushroom or Dhingri' in India is relatively new to the mushroom industry. *Pleurotus pulmonarius* is not cultivated in the state of Odisha very frequently. Hence, cultivation technology was standardized to incorporate P. pulmonarius mushroom in the oyster mushroom cultivation in Odisha along with the other dominant species. Four different substrate pasteurization methods along with a control (untreated) were evaluated for their yield potential. The results showed that steam treatment of substrate gave highest mushroom yield (BE 98.90%) and vield obtained in chemically treated substrate (BE 96.63%) was statistically at par with the steam treatment. But in chemically treated substrate incubation period was longer (17.50 days) and crop duration was 24.25 days. The untreated substrate (control) produced the lowest yield (BE 79.28%). Nine organic supplements including control were evaluated for their influence on yield and yield attributing parameters of *Pleurotus pulmonarius*. The biological efficiency was reported numerically highest (93.37%) in boiled wheat which was at par with the yields realized from chicken manure (93.07%), boiled maize (92.53%), rice bran (92.33%), maize meal (91.10%), untreated control (91.07%), and Bengal gram powder (90.99%). Among the six diverse bag dimensions, 30×25 cm bag accommodating 1500g substrate was found superior in respect of biological efficiency.

Keywords: Oyster mushroom, P. pulmonarius, substrate treatment, supplementation

Pleurotus mushroom, generally referred to as 'Oyster mushroom or Dhingri' in India is relatively new to the mushroom industry but has gained popularity at a tremendous pace and today it is cultivated in about 25 countries of far-East Asia, Europe and America. It is the third largest cultivated mushroom in the world and contributing around 16% of the total world production (Singh *et al.*, 2021) while China alone contributes 88% of the world production. The other major oyster mushroom producing countries are South Korea, Japan, Italy, Taiwan, Thailand, Philippines, etc. At present, India produces only small

Orissa, Karnataka, Maharashtra, Andhra Pradesh, Tamil Nadu, Bihar, Madhya Pradesh, Chhattisgarh, Jharkhand, West Bengal and in the North-Eastern states of Meghalaya, Manipur, Mizoram, Tripura and Assam (Sharma, *et al.*, 2017).

quantities (21,272 tonne) of oyster mushroom in

In India, Odisha is one of the leading states in terms of oyster mushroom production. Different oyster species are under cultivation in the state with annual production of 8000 metric tonnes contributing to about 40% of the total mushroom production of the state.

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Most of the agro-climatic conditions in Odisha are suitable for growing oyster mushroom from month of July to February comprising of the rainy and winter season. However, people of Odisha prefer growing oyster mushroom during winter season i.e. from November to February when productivity of paddy straw mushroom declines owing to low temperature. *Pleurotus florida* and *Pleurotus sajor-caju* are the preferred species in the state. Besides these two species, *P. eous* is also gaining popularity throughout the state.

The oyster mushroom is cultivated under thatched roof (indoor cultivation) in both coastal and inland districts. Cultivation is done largely on non pasteurized paddy straw substrate without organic supplements. The yield obtained varies betweeen 66-100 kg per 100 kg dry straw. The reasons for such wide variations in the level of productivity are attributed to use of nonpasteurized straw without supplements and lack of aftercare. But there is scope for yield increase by effective substrate management besides using productive strains of the species. Quantity of substrate/bags under appropriate environmental conditions is also important in production of mushroom.

P. pulmonarius could well be incorporated in to the mushroom farming system during winter season in Odisha besides the other *Pleurotus* species, as this species was not under cultivation earlier. Hence the effective of various conventional as well as nonconventional substrates and various growing techniques need to be standardized. Therefore, an attempt has been made to investigate the suitable package of practices for *P. pulmonarius* cultivation.

MATERIALS AND METHODS

The present investigations were undertaken to study the effect of the pasteurization methods, organic supplements (additives) and quantity of the substrate/ bag on the production of oyster mushroom (*P. pulmonarius*).

Substrate pasteurization methods used

A total of four pasteurization methods along with an untreated control were evaluated in this investigation. In treatment 1, chopped and moistened substrate was soaked in hot water (70°C) for one hour. Excess water was drained and spawn was added @ 10% of the dry weight. In treatment 2, pre-wetted chopped straw was steam pasteurized at 65-70°C for 4-6 hours, cooled and spawned. In treatment 3, moistened straw was spread on cemented floor in thin layer and covered with clean polythene sheet (100 gauges) and treated in sun-light from 10.00 AM to 4.00 PM in bright sunny days. In treatment 4, the substrate was chemically pasteurized using formaldehyde and Carbendazim (100 litre of water, 7.5 g bavistin and 125 ml 40% formaldehyde). Excess water was drained and spawned. Chopped straw soaked in clean and cold water served as control. Each treatment was replicated four times. Appropriate conditions were maintained in the cropping room for spawn run and induction of fruit bodies. Observations on time taken for spawn run, days taken for pinhead formation, time taken for 1st and 2nd flush, average fruiting body weight and yield of mushroom (biological efficiency) were recorded.

Organic supplements used

A total of nine organic supplements i.e. maize powder, ragi powder, Bengal gram powder, mustard oil cake, chicken manure, boiled wheat, rice bran, tapioca and boiled maize were evaluated along with a control (no supplement) to see their effect on productivity of *P. pulmonarius*. The supplements were used @ 200g per bag for all sizes. The supplements were autoclaved for 20 minutes at 10 psi pressure and cooled down to room temperature before adding to the substrate. The treatments were replicated thrice. Observations on time taken for spawn run, time taken for first harvest, average fruiting body weight and mushroom yield (biological efficiency) were recorded.

Quantity of substrate used

Effect of quantity of substrate/bag on the productivity of P. pulmonarius was investigated during the study. Various quantities of dry substrates (500g, 1000g, 1500g, 2000g, 2500g and 3000g) were used for preparation of bags. Layer spawning was followed with the pre-wetted and pasteurized substrate at the rate of 10% of the dry weight in bag of variable sizes, tied and incubated. Each treatment was replicated four times. The bag containing 1500g of dry substrate served as check. Appropriate light, temperature, humidity and substrate moisture was maintained in the cropping room. Observations on time taken for spawn run, first harvest, average weight of fruit body and mushroom yield were recorded. After the bags were colonized by the fungus, they were shifted to the cropping room for induction of fruiting.

Spawning of bags

Freshly prepared grain spawn (20-30 days old) was used for spawning. Spawning was done in a prefumigated room (48 hours with 2 % formalin) @ 10 % of the dry weight of the substrate (150 g of spawn per bag having 1.5 kg of dry substrate). The spawn was divided into four parts and layered spawning was done in polythene bag of 80cm x 40 cm size. The bags were tied up and ten to 15 small holes (0.5-1.0 cm dia) were made on all sides of the bag including two to four holes in the bottom to facilitate leaching out of excess water and ventilation.

Fruit body induction and harvest

The colonized bags were arranged on wooden shelves with a minimum distance of 15-20 cm between two bags. Appropriate light (200 lux for 8-12 hours a day), temperature (20-25°C) and relative humidity (75-80 %) were maintained to facilitate fruiting. The bags were sprayed with water twice daily to maintain moisture status of the substrate. Three to four days after opening of bags, mushroom primordia started to appear. Fruit bodies were harvested in about three days after their appearance. Three flushes were harvested at an interval of 7-10 days.

Yield and biological efficiency

The biological efficiency was calculated by the following formula

BE (%):
$$\frac{\text{Fresh weight of mushroom harvested}}{\text{Dry weight of the substrate}} \times 100$$

RESULTS

Effect of substrate pasteurization methods on mushroom productivity

Data recorded on days to spawn run, days to first harvest, average fruit body weight (g) and biological efficiency (%) are presented in Table 1. Steam treatment of substrate at 60-80°C for four hours was found superior in terms of days to spawn run (14.50d), days to first harvest (21.75d) and biological efficiency (98.90%). Chemical treatment of the substrate was found to have longer incubation period (17.50d) and crop duration (24.25d). However, the average fruit body weight (9.15 g) was better in chemically treated substrate than the steam treated substrates. The untreated substrate produced the lowest yield (79.28 %). The steam treatment was thus better in terms of yield of mushroom.

Influence of organic supplements on biological efficiency of *P. pulmonarius*

The experiment was designed to evaluate as number of organic additives i.e. maize meal, ragi powder, bengal gram powder, mustard oil cake, chicken manure, boiled wheat, rice bran, tapioca pearls and boiled maize for their effect in improving mushroom productivity over the untreated check (control). Data recorded on time taken for spawn run (d), first harvest (d), average weight of fruit bodies (g) and biological efficiency (%) are presented in Table 2.

Sl. No.	Treatment	Days to spawn run	Days to first harvest	Average fruit bodies wt. (g)	Biological efficiency (%)
1	Boiled water treatment	16.00	24.00	7.88	91.80
2	Steam treatment	14.50	21.75	8.45	98.90
3	Substrate solarisation	16.50	24.25	8.80	82.95
4	Chemical treatment	17.50	24.25	9.15	96.63
5	Control (No treatment)	16.50	22.50	7.10	79.28
CD (0.05)	-	1.07	1.12	1.25	6.29
CV (%)	-	7.48	3.10	9.83	4.54

Table 1. Effect of substrate pasteurization methods on biological efficiency of P. pulmonarius

Table 2. Effect of Organic supplements on biological efficiency of P. pulmonarius

Sl. No.	Treatment (Organic supplements)	Days to spawn run	Days to first harvest	Average fruit bodies wt. (g)	Biological efficiency (%)
1	Maize meal	17.67	24.33	10.33	91.00
2	Ragi powder	18.67	26.67	7.37	78.37
3	Bengal gram powder	17.33	23.33	10.20	90.99
4	Mustard oil cake	19.00	25.67	7.17	79.00
5	Chicken manure	18.33	25.00	10.93	93.07
5	Boiled wheat	17.67	24.33	11.00	93.37
7	Rice bran	17.33	24.67	10.94	92.33
8	Tapioca pearls	18.00	25.67	7.08	80.40
)	Boiled maize	17.00	23.33	9.02	92.53
10	Control (No supplement)	17.33	23.33	10.01	91.07
CD (0.05)	-	1.29	2.01	0.98	5.42
CV (%)	-	4.22	4.76	6.05	3.58

Analysis of data indicated less variation among the incubation period of the test species under the influence of nine additives. It ranged from 17 days in boiled maize to 19 days in mustard oil cake. The days to first harvest varied from 23.33 days to 26.67 days in the investigation. The average fruit body weight (g) was observed to be higher in treatment with wheat grain as supplement (11.00 g) statistically at par with rice bran (10.94 g), chicken manure (10.20 g), maize meal (10.33g) and Bengal gram powder (10.20g). The biological efficiency was recorded numerically highest (93.37%) in boiled wheat grain supplement that was at par with the yields realized in chicken manure (93.07%), boiled maize (92.53%), rice bran (92.33%), untreated control (91.07%), maize meal (91.00%) and Bengal gram powder (90.99%).

Influence of post-spawning practices on biological efficiency of *P. pulmonarius*

Diverse post-spawning practices such as uncovering the oyster bags just after mycelia colonization or maintaining the bags throughout the crop period either with small holes (0.5 cm) or big

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holes (1.0 cm) are in operation among the growers. Hence, an attempt was made to assess their yield performance during the winter season of 2015-16. Data recorded on days to spawn run and first harvest, number of fruit bodies per bag, average fruit body weight (g) and biological efficiency (%) were analyzed and are presented in Table 3.

Data indicated that the days to spawn run was statistically equal in all the three treatments ranging from 15.67-16.00 days. However, the days to first harvest (crop duration) was significantly lowest (21.50) and number of fruit bodies was significantly more (183.33) in the uncovered bags. Average fruit body weight was recorded lowest (7.58 g) in uncovered bags. However, they are recorded at 9.98 g and 9.73 g in bags with 1.0cm and 0.5cm holes, respectively statistically at par with each other. The biological efficiency was found highest in uncovered bags (97.34%), which were at par with bags with 1.0 cm holes (96.47%). Bags with smaller holes were associated with a biological efficiency of 91.68 percent. Hence, it was ascertained that bags with either big holes of 1.0 cm size or uncovered ones were almost parallel in their yield performance as recorded in the investigation.

The investigation on the effect of bag dimension on mushroom productivity indicated the superiority of 30x25 cm bag accommodating 1500g dry substrate with significantly higher yield of 101.18 percent among all the six size of bags evaluated. It was closely followed by the bag dimension of 25x20 cm accommodating 1000g dry substrate with 98.40% biological efficiency. It was observed that the large sized bags (40x25 cm, 50x25 cm and 60x25 cm) were poor yielders (BE 87.40-90.05%). The bag size of 20x15 cm accommodating 1000 g substrate could give 95.60% biological efficiency which would probably be the best option for small growers.

DISCUSSION

Pleurotus is relatively new to the mushroom industry but has gained popularity at a tremendous pace because of the simple cultivation technique, high productivity, choice of species, adoptability to variety of substrate and above all longer shelf life. Out of 38 species described under the genus, about 25 species are commercially cultivated in different parts of the world.

In Odisha, two species namely, *Pleurotus sajor-caju* and *Pleurotus florida* are popular among the farmers owing to their high yield potential and acceptable taste and flavour. However, efforts are being made to popularize few more promising species like *P. pulmonarius* This species has got high yield potential, besides having excellent aroma and good shelf life. An attempt has by made to develop the package of practices for commercial cultivation of *P. pulmonarius* in this investigation.

The mycelia of *Pleurotus* mushroom can grow on simple water treated straw but number of competitor moulds may restrict the growth of mycelium. Thus substrate pasteurization becomes very important to get a good crop at commercial level.

Sl. No.	Treatment (Post-Spawning practices)	Days to spawn run	Days to first harvest	No. of fruit bodies/kg	Average fruit bodies wt (g)	Biological efficiency (%)
1	Bags with 1.0 cm hole	15.83	24.17	146.83	9.98	96.47
2	Bags with 0.5 cm hole	16.00	24.00	144.00	9.73	91.68
3	Bags uncovered	15.67	21.50	183.83	7.58	97.34
	CD (0.05)	1.17	1.36	6.98	1.18	4.07
	CV (%)	5.77	4.56	3.43	10.06	3.33

Table 3. Influence of Post-Spawning practices on biological efficiency of P. pulmonarius

Four different substrate pasteurization methods were evaluated for their yield potential and steam treatment was found to be most suitable with a mushroom yield (98.90%). The steam penetrates better in the substrate and pasteurization becomes more effective (Siqueira *et al.*, 2012). Besides, the chemical treatment leaves residues in the substrate, which probably lengthened the incubation period and the crop duration initially. The performance of chemical treatment was in agreement with the findings of Vijay and Sohi (1987).

The nitrogen contents in most of the substrates range between 0.5-0.8% and hence addition of organic nitrogen in the substrate helps in obtaining higher yields. Some of the common supplements in use are wheat bran, rice bran, cotton seed mill, mustard oil cake, groundnut cake, soybean cake, boiled wheat grain, chicken manure, maize powder, etc. They are used at 5-10% on dry weight basis of the substrate for yield improvement in oyster mushroom. In the present investigation, nine additives along with control (no additives) were evaluated for their yield potential. The data indicated that minimum incubation period in boiled maize. The crop duration was found lowest in Bengal gram powder. The biological efficiency was found maximum in boiled wheat. Various workers have evaluated diverse groups of supplements for yield enhancement of Pleurotus. Upadhyay et al. (2002) indicated that rice bran at 5% level was the appropriate substrate for *Pleurotus*. Veena et al. (1994) evaluated supplements in combination and indicated that Pleurotus florida performed well with a combination of rice bran and soys dal powder. Pleurotus mushroom have an appreciable degree of competitive saprophytic ability and therefore, the bags receiving no additive also produced statistically equal yield with the boiled wheat supplementation.

Bag dimension and quantity of substrate play a vital role in maintaining moisture and temperature and there by influencing the sporophore yield to a great extent. Suman and Sharma (2007) recommended use of 60x45 cm bags of 125-150 gauze thickness for oyster mushroom cultivation, which accommodated 3.0 kg of dry substrate. However, this investigation ascertained that medium sized bags (30x25 cm) with 1500g straw could be the best option for commercial growers.

CONCLUSION

From the experiments, it could be concluded that among the pasteurization methods of substrate for *P*. *pulmonarius*, steam treatment was found to be the best. The study on supplementation of substrate indicated that all the tested supplementations except ragi powder, mustard oil cake and tapioca pearls were statistically at par in respect of yield performance. The finding showed that oyster mushroom could successfully be grown in the absence of organic supplements also. With respect to bag size and quantity of the substrate, the bag size of 30 x 25 cm accommodating 1500g substrate gave the maximum biological efficiency.

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