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Research Article

EFFECT OF ORGANIC SUPPLEMENT ON OYSTER MUSHROOM SPAWN PRODUCTION

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ABSTRACT

The cultivation of mushrooms is a biotechnological intervention for the conversion of various lignocellulose agro-wastes into proteins. Jharkhand and its neighbourhood are potential area for oyster mushroom production due to its favourable agro-climate, plenty of agro-wastes and *Hypsizygus ulmarius* good awareness and liking for mushroom consumption. They also provide other much needed nutrients like Essential Amino Acids (EAA), minerals and vitamins and keep us fit and healthy. Oyster mushroom cultivation in India is very popular among the farmers due to its simple growing technique and the availability of its good number of species adapted to wide range of temperature - from tropical to temperate. *Pleurotus florida* exhibits adaptability to perform well in sub-tropical areas and hence it is more popular in Chotanagpur Plateau in general and Ranchi District in particular. In the current study, therefore, *P. florida* was selected as a test species. Experiments on the species of Oyster mushroom were designed with the sole objective to reduce the input-cost of their cultivation by promoting the use of cheaper recycled agro-wastes like vermi-compost for spawn media preparation. The study concluded that supplemented spawn medium like Wheat (50%) + Vermi Compost (50%) and Wheat (75%) + Vermi Compost (25%) gave better result as well as its performance is at par with 100% wheat for the spawn production of *P. florida* and. So this can be recommended to spawn production laboratories for the use of this methodology for reducing the cost of spawn production. Vermi-compost, which is a waste-recycled material, has been identified as a potential substitute for cereal grains. Their replacement (partial or total) with Vermi-compost has opened the doors to make mushroom farming a complete recycling process, and also to achieve organic farming of mushrooms a reality sooner than later. Therefore, more such studies on other edible domesticated mushroom species need to be made in future.

Keywords: Oyster mushroom, Spawn production, Jharkhand.

1. INTRODUCTION

Mushrooms along with other fungi are living things which are neither plant nor animal. Consumption of edible fungi, especially the mushrooms, to satisfy the protein hunger of the vast human population of that time might prove as a potent and notable step in the history of mankind [1]. They have been placed in a kingdom of their own, called Myceteae. Mushrooms are macro fungi with a distinctive fruiting body, which can be either epigeous or hypogeous and large enough to be seen with naked eye and to be picked by hand. The most common type of mushrooms is umbrella shaped with a pileus (cap) and a stipe (stem), that is, *Lentinula edodes* (Shiitake). Other species additionally have a volva (cup), that is, *Volvariella volvacea*, or an annulus (ring), that is, *Agaricus campestris*, or both, that is, *Amanita muscaria*.

Jharkhand and its neighbourhood are potential area for oyster mushroom production due to its favourable agro-climate, plenty of agro-wastes and good awareness and liking for mushroom consumption. However, earlier mushroom worker has reported that Green Mould, Aspergillus and bacteria commonly infects the oyster beds in this area [2]. The genus Trichoderma, commonly called as weed mould in mushroom industry is responsible for causing Green mold disease of mushrooms. Also, some earlier attempts have shown the

potentials of botanicals and Neem-oil formulations in reducing Trichoderma infection. Cultivation of specialty mushrooms on lignocellulosic wastes represents one of the most economically and cost-effective organic recycling processes. The technology can also limit air pollution associated with burning agriculture wastes as well as to decrease rodents, pests and deleterious fungal inoculum populations [3]. In any mushroom cultivation programme the primary requisite in preparing a suitable spawn. Mushrooms can grow on a variety of fresh lignocellulosic residues requiring very little pretreatment [4].

2. MATERIAL AND METHODS

The present study was conducted in the Mushroom Laboratory of the Ramakrishna Mission Vivekananda University, IRTDM faculty Centre, Morabadi, Ranchi (Jharkhand). The experiments of the project were conducted with master cultures of *BacilllusNakamurai*, *Hypsizygus ulmarius* and *Pleurotus florida,* obtained from the Culture Bank of Ramakrishna Mission Vivekananda University, Faculty Centre- IRTDM, Ranchi, Jharkhand and Directorate of Mushroom Research Centre, Solan, HP.

2.1. Culture media for isolation and maintenance of pure cultures of test fungi

Potato dextrose agar medium was prepared as per the standard procedure.

2.2. Preparation of vermi-compost

For experiment Vermi-compost were collected from Divyayan Krishi Vigyan Kendra (DKVK), Ramakrishna Mission Ashrama, Morabadi, Ranchi, Jharkhand. The preparation process of Vermi-compost was as follows: Vermi-composting is a process of converting organic wastes like agricultural residue, leaf litter, and kitchen wastes into Vermi-compost through the action of earth worm species. In DKVK (Ranchi), they utilize *Eisenia foetida* (earth worm sp.) for production of Vermi-compost. The most pit size is 2m×1m×0.75m; the bottom layer is filled with bedding material (dry leaves) with a thickness of 2.5m to 5m. Then filled with partially decomposed cow dung, water is sprinkled on it there after cocoons or earth worms are introduced on to the bed. If the dung is unavailable, a little amount of Vermi-compost or small amount soil into the bed. The moisture level must be 50% to 60%. It will take 2 to 3 weeks to complete the formation of Vermi-compost.

2.3. Modified method of spawn preparation to determine the moisture level of wheat grains to be used for spawn medium

At first 100 gm of wheat grains were taken and kept in hot air oven at 120°C temperature and at every 20 minutes interval, the weight of wheat grains was recorded. When the weight of grains became constant; it was taken as the net weight of grains for calculating the Initial moisture content of the wheat grains, using the following formula: -

$$\text{Moisture Level Per cent (\%)} = \frac{(Wi - Wn)}{100} \times 100$$

Where,Wi= Initial Wt. of wheat grains taken, Wn= Final Wt. of wheat grains taken

2.4. Preparation of grain spawn medium

Good quality of wheat free from broken, insect and pest damage mould infestation was taken. Moisture quality of grains was checked. 2 kg of wheat grain were soaked in water for 10 to 20 minutes and washed to remove any dirt from the grains. The washed grain was treated with well guard dilution (10-5 dilution) to submerge the grain and maintain moisture at 55 to 60 per cent. The grains were soaked in well-guard dilution for 18 to 24 hrs. The grains during soaking were covered with polythene cover to prevent the moisture loss from the wheat grains. The grains were taken out and spread on the thin polythene sheet, shaded to dry out the water sticking on the grains. Then calcium carbonate; 2% (20 grams per kg), was added and mixed properly with grains.

2.5. Preparation of vermi-compost spawn medium

Vermi-compost was taken and sieved to remove any clogs, earth worms or any sticks and any other unwanted materials. Moisture level of vermi-compost was checked. Moisture level must be maintained between 50 to 60 per cent. If the moisture level is less than 50 to 60 per cent thereafter sufficient moisture was added and soaked for 1 hour mix 6% of gypsum (60 grams per kg).

2.6. Treatment

2.6.1. Control (100% Wheat Grains)

The grain spawn medium were filled in the glass bottles and filled up to their 2/3 capacities and plugged them with jute cotton covered with polypropylene paper and wrapped with rubber. The spawn bottles were autoclaved at 15 Ib psi pressure for at least 45 minutes and were shaken properly to avoid clump formation after

taking them out, then transferred to the inoculation chamber (laminar air flow) for overnight cooling before using them for inoculation.

2.6.2. Wheat grains (50%) + vermi-compost (50%)

Spawn bottles were filled with vermi-compost and wheat grains (50:50 percentage compositions on the basis of the experiment). Thus, the prepared media was filled with half litre glass saline bottles up to their 2/3rd capacities before plugging them with jute cotton covered with polypropylene paper tied with rubber band. The spawn bottles were autoclaved at 15 Ib psi pressure for at least 45 minutes and were shaken properly to avoid clump formation after taking them out, then transferred to inoculation chamber (laminar air flow) for cooled overnight before using them for inoculation.

2.6.3. Wheat grains (75%) + vermi-compost (25%)

Preparation of wheat grain and vermi-compost material are similar as above mentioned. In these processes the spawn bottles were filled with 75 % wheat grain and 25% Vermi-compost on the basis of the experiment. Then prepared media was filled with half litre glass bottles up to 2/3 capacities and plugged with jute cotton and jute cotton wrapped with polypropylene paper tied with rubber band. Thus, the spawn bottles were autoclaved at 15 Ib psi pressure for 45 minutes. Then bottles were transferred for further cooling.

2.6.4. Vermi-compost (75%)+ wheat grains (25%)

A ratio of 75% of vermi-compost and 25 % of wheat grains was filled in 12 glass bottle up to 2/3 capacity and plugged with jute cotton and wrapped with polypropylene paper tied with a rubber. The bottles were autoclaved at 15 Ib psi pressure for 45 minutes. Further the bottles were transferred into inoculation chamber for further cooling.

2.7. Inoculation of grain spawn-bottles

For inoculation of medium spawn bottles, fresh inoculums were prepared in several petri plates. Plates showing mycelium growth almost fill in the plates were used for obtaining 1mm mycelial cubs from edges of the growing cultures *P. Florida* and *Hypsizigus ulmarius* for using them as inoculate. The sterilized spawn bottles were then inoculated with inoculum under completely sterilized conditions. Then the spawn bottles were completely placed in the incubator at 22±2°C in BOD incubators till the grain medium was fully impregnated with mushroom mycelium. However, the bottles were shaken at regular intervals to enhance and ensure a uniform growth of the mushroom mycelium.

2.8. Statistical analysis of the data

The data obtained were quantified as per rules of statistical analysis for drawing meaningful conclusions. A few statistical techniques employed in various phases of the present study are Variance, Standard Deviation, Co-efficient of Variance, Completely Randomized Design, Critical Difference.

3. RESULTS AND DISCUSSION

Oyster mushroom cultivation in India is very popular among the farmers due to its simple growing technique and the availability of its good number of species adapted to wide range of temperature - from tropical to temperate. *Pleurotus florida* exhibits adaptability to perform well in sub-tropical areas and hence it is more popular in Chotanagpur Plateau in general and Ranchi District in particular. In the current study, therefore, *P. florida* was selected as a test species. Experiments on the species of Oyster mushroom were designed with the sole objective to reduce the input-cost of their cultivation by promoting the use of cheaper & recycled agro-wastes like Vermi-compost for spawn media preparation.

3.1. Growth- pattern of Pleurotusfloridaspawn on supplemented spawn media

This experiment was conducted on the following media - combinations

T1 = Wheat Grains (50 %) + Vermi-Compost (50 %),
T2 = Wheat Grains (75%) + Vermi-Compost (25%),
T3 = Wheat Grains (25%) + Vermi-Compost (75%),
T4 = Wheat Grains (100%) and
T5 = Vermi-Compost (100 %)

A perusal of data recorded in Table 1spawn mycelial growth of the fungus *P. florida* on different spawn production substrate like Wheat Grains (50 %) + Vermi-Compost (50 %), Wheat Grains (75%) + Vermi-Compost (25%), Wheat Grains (25%) + Vermi-Compost (75%), Wheat Grains (100%) and Vermi-Compost (100 %) gives significant effect and in these all treatment, T1 which was Wheat Grains (50 %) + Vermi-Compost (50 %) gives better result than rest of other our treatments.

Table 1: Linear spawn growth of *Pleurotus florida* on different supplemented spawn medium

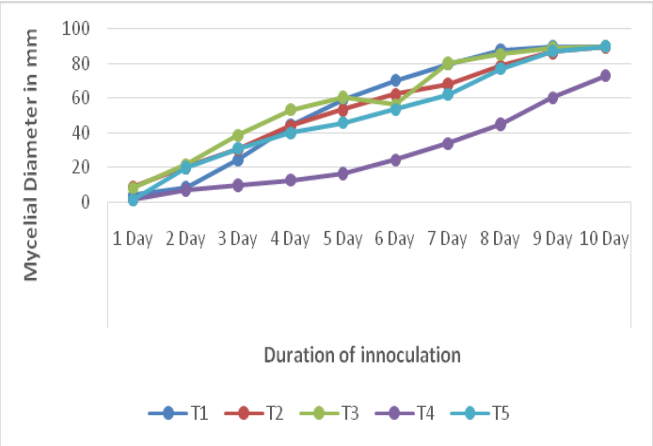
	Mycelial Diameter in mm						
Dilutions	T1	T2	T3	T4	T5	S.Em	CD 5%
1st day	4.00	9.00	8.40	1.40	1.40	11.95	29.14
2nd day	8.40	20.20	21.8	7.00	19.80	20.78	50.69
3rd day	24.20	30.80	38.80	9.80	31.00	25.40	61.95
4th day	44.60	44.40	53.60	12.60	40.00	25.72	62.75
5th day	59.00	53.40	60.60	16.60	46.00	28.68	69.96
6th day	70.40	62.20	56.40	24.40	53.60	39.29	95.84
7th day	80.00	68.20	80.40	33.80	62.20	28.63	69.83
8th day	88.00	78.60	85.60	45.00	77.20	26.72	65.17
9th day	90.00	86.80	89.20	60.40	87.40	21.61	52.70
10th day	90.00	89.60	90.00	73.00	90.00	13.47	32.86

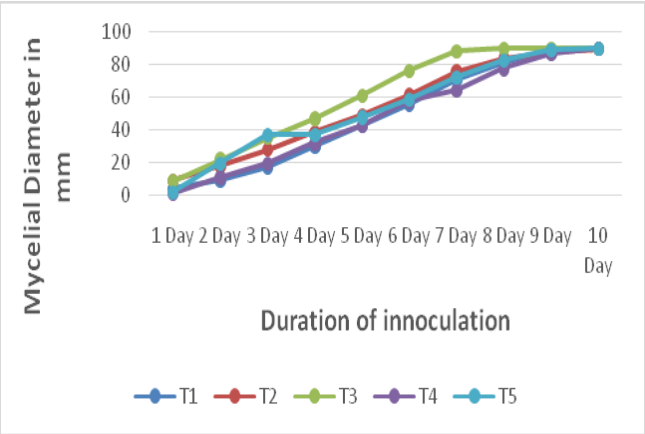
T1 =Wheat Grains (50 %)+ Vermi-Compost (50 %), T2 =Wheat Grains (75%) + Vermi-Compost (25%), T3 =Wheat Grains (25%) + Vermi-Compost (75%), T4 =Wheat Grains (100%), T5 =Vermi-Compost (100 %)

Table 2: Linear spawn growth of Hypsizigusulmarious on different supplemented spawn medium

	Mycelial Diameter in mm						
Dilutions	T1	T2	T3	T4	T5	S.Em	CD 5%
1st day	4.00	9.00	8.40	1.40	1.40	11.95	29.14
2nd day	8.80	17.80	21.60	11.00	18.80	20.94	51.07
3rd day	16.60	27.60	34.80	19.20	37.00	22.13	53.98
4th day	29.80	38.60	46.80	32.00	37.00	19.99	48.77
5th day	42.80	48.80	61.00	42.60	47.60	19.90	48.54
6th day	55.80	61.20	76.40	57.60	58.60	16.70	40.74
7th day	70.80	76.00	88.60	64.60	72.60	16.08	39.21
8th day	81.20	84.00	90.00	77.60	83.00	12.73	31.06
9th day	88.80	87.80	90.00	87.00	89.20	6.39	15.60
10th day	90.00	89.40	90.00	90.00	90.00	1.20	2.93

T1 =Wheat Grains (50 %)+ Vermi-Compost (50 %), T2 =Wheat Grains (75%) + Vermi-Compost (25%), T3 =Wheat Grains (25%) + Vermi-Compost (75%), T4 =Wheat Grains (100%), T5 =Vermi-Compost (100 %)





The graph also shows that all treatments gives significant effect than the control.

A perusal of data recorded in table 2 spawn mycelial growth of the fungus *Hypsizigus ulmarious* on different spawn production substrate like Wheat Grains (50 %) + Vermi-Compost (50 %), Wheat Grains (75%) + Vermi-Compost (25%), Wheat Grains (25%) + Vermi-

Compost (75%), Wheat Grains (100%) and Vermi-Compost (100 %) gives significant effect and in these all treatment T3 which was Wheat Grains (25 %) + Vermi-Compost (75 %) gives the better result than rest of other our treatments.

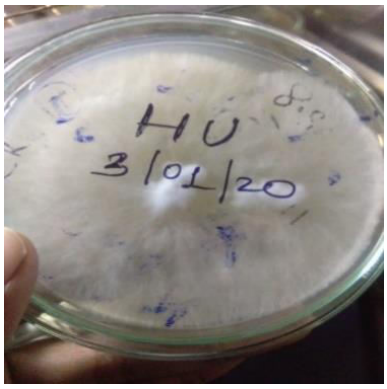
This graph also shows that all treatments gives significant effects.

From this experimental project we found that supplemented spawn medium like Wheat (50%) + Vermi Compost (50%) and Wheat (75%) + Vermi Compost (25%) is giving better result as well as its performance is at par of 100% wheat for the spawn production of *P. florida* and *H. ulmarius*. So that we can recommend spawning production labs for using this methodology for reducing the cost of spawn production. The findings in this study are in agreement with previous findings that substrates used in mushrooms cultivation have effect on chemical characteristics of mushrooms [5] indicating a variation in the capability of such substrates to support mushroom growth [6].

Thein the rate of growth variation, spawn run and total yield on the different substrates in this study validates the findings of Nasir et al. (2012) [7] in the use of sawdust of different woods for the cultivation of oyster mushroom (*P. ostreatus*). Ingredients with high content of organic cabon compared to protein-rich components, such as agricultural and commercial waste products, are cheaper and readily available in local producing areas [8].

The quality of oyster mushroom *P. florida* depends on the length of stalk; the higher the length of stalk, the poor the quality of mushroom [9]. Vermi-compost, which is a waste-recycled material has been identified as a potential substitute for cereal grains mostly wheat. Their replacement (partial or total) with vermi-compost, a well cherished goal has both monetary and ecological advantages [10].

It has opened the doors (i) to make mushroom farming a complete recycling process, and also (ii) to achieve healthy organic farming of high quality mushrooms a reality sooner than later. The two systems of organic farming and recycling will result in a product that is more stable and homogenous [11].



Prepared Pure Culture of Mushroom Spp.

Mixing of $CaCo_3$ in Wheat

Mixing of Wheat and vermi compost in different ratio.

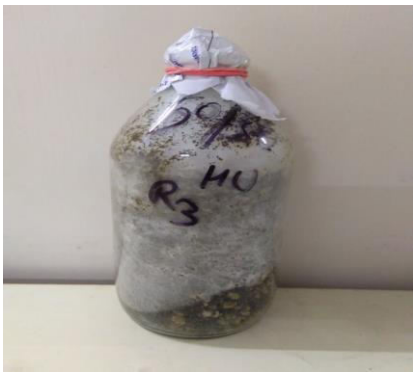
Showing growth of *P. florida*on Wheat (25%) and Vermi Compost (75%)

Showing growth of *P. florida* onWheat (50%) and Vermi Compost (50%)

Showing growth of *P. florida*onWheat (75%) and Vermi Compost (25%)

Showing growth of *P. florida*onVermi Compost (100%)

Showing growth of *P. florida*onWheat (100%)



Showing growth of *HU in*Wheat (50%) and Vermi Compost

Reports support that mushrooms can play a pivotal role in bioremediation of polyethylene with the aid of the extracellular enzymes such as laccases and manganese peroxidase. Mycelium of *Pleurotus florida* are able to utilize the polyethylene sheet as a carbon source for the growth and can play a role in bioremediation and biodegradation of low density polyethylene. The product could meet the pathogen reduction requirements. Therefore, more such studies on other edible domesticated mushroom species need to be made in future.

4. CONCLUSION

The findings of this investigation lead us to conclude that the primary goal of this study to reduce the input-cost of oyster mushroom spawn production has been achieved to a large extent. Further investigations are necessary to determine the influence of other growth promoting additives.

Conflict of interest
None declared

Source of funding
None declared

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