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Evaluation of different substrates for spawn production of white oyster mushroom (*Pleurotus florida*)

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Abstract
Mushrooms are considered as the important constituent of the human's diet due to their high nutraceutical value. The mushroom spawn is the basic material for the production of quality mushrooms. The present study was carried out to evaluate the different substrates for spawn production of white oyster Mushroom (*Pleurotus florida*). Seven locally available substrates *viz*., maize cobs, maize seeds, wheat grains, chickpea grains, walnut shells, almond chips and barley grains (check) were evaluated for their suitability for the spawn production. Results revealed that walnut shells and almond chips proved to be more efficient substrates with regards to spawn run, spawn texture and pH. The spawn run in walnut shells and almond chips was completed in 6.75 days and 7.00 days, respectively as compared to 7.25 days in barley grains (check). The spawn colour and textures on different substrate showed slight variation, while the spawn colour ranged from 'brownish-white' in walnut shells and almond chips, 'light brown' in chickpea grains to 'white' in other grain substrates, Spawn texture ranged from 'Cottony' to 'Fluffy'. Cottony texture was recorded in chickpea grains, while as in all other substrates 'fluffy' texture was observed. pH of the spawn ranged from 6.65 in walnut shells to 7.95 in maize shelled cobs. All the substrates were found suitable for spawn preparation of *Pleurotus florida*.

Keywords: Nutraceutical value, substrates, spawn production, efficient, colour

Introduction
Mushrooms are recognized as important food products and their usage is increasing day by day because of their tremendous role in human health, Nutritional and therapeutic use (Patrick *et al.*, 2011) [10]. Oyster mushroom is the second important commercially grown mushroom in India after button mushroom. Oyster mushrooms are the easiest and least expensive commercial mushrooms to grow since they are well known for transforming crop residues into food proteins and are considered as a potential source of revenue, alternative food production, work provision and agricultural waste recycling (Banik and Nandi, 2004) [2]. In oyster mushrooms, the high content of nitrogen and protein increases biological productivity and yield while reducing the growth time.

Oyster mushroom is an edible mushroom of excellent taste and flavor (Mondal *et al.*, 2010) [6]. Oyster mushroom cultivation is prevalent due to low-cost technology and the simple availability of various substrates for its cultivation. It is widespread in temperate, subtropical and tropical regions and displays the highest yield among the genus *Pleurotus* (Kong, 2004) [5]. *Pleurotus florida* contains medicinal and pharmacological metabolites such as antimicrobials, immunostimulants, antioxidants and anti-tumour medicines. (Elmastas *et al.*, 2007, Israilides *et al.*, 2008) [3, 4].

Cultivation of *Pleurotus florida* gives a wide range of response to different agro-wastes. In view of low cost production methods, strong economic returns, attractive characteristics and obvious potentialities of *Pleurotus florida* oyster mushroom.

Oyster mushrooms are commonly grown on rice straw or wheat straw. They can, however, be grown on a wide variety of ligno cellulosic substrates, thus playing an important role in the management of organic waste which may otherwise be problematic in disposal. Keeping in mind the environment and agricultural residues available in the area, there is ample scope for its commercial production (Alice and Kustudia, 2004) [1].

Spawn comprises mycelium of the mushroom and a supporting medium which provides nutrition to the fungus during its growth. The propagating material used by the mushroom growers for planting beds is called spawn.

The spawn is equivalent to vegetative seed of higher plants (Pathak *et al*., 2000) [8]. In mushroom growing technology, the inoculums are known as the 'spawn'. Spawn is a medium that is impregnated with mycelium made from a pure culture of the chosen mushroom strain. Spawn production is a fermentation process in which the mushroom mycelium will be increased by growing through a solid organic matrix under controlled environmental condition.

Materials and Methods

The present study was carried out at Mushroom Technology and Training Centre, Division of Plant Pathology, Sher-e-Kashmir University of Agricultural Sciences and Technology of Kashmir, Faculty of Agriculture, Wadura during 2020. Seven commonly available substrates *viz.*,, wheat grains, maize grains, chick pea grains, maize cobs, walnut shell, almond chips and barley grains (check) were evaluated by adopting the procedure given by Patil (2012) [9] for spawn preparation.

Procurement and maintenance of culture

The pure culture of *Pleurotus florida* used in present investigation was procured from Directorate of Mushroom Research, Chambaghat, Solan. The culture was multiplied further in tubes containing potato dextrose agar medium. Apparently healthy, unbroken wheat grains were used for 'Master Spawn' preparation and the procedure given by Patil (2012) [9] was followed.

Evaluation of substrates for spawn production of *Pleurotus florida*

Adequate quantities of following substrates (Table 1) were cleaned and after removing the admixture evaluated for spawn production.

Table 1: Substrates evaluated for spawn production of *Pleurotus florida*

Vernacular name	Botanical name	Plant parts used
Wheat (check)	*Triticum aestivum*	Grains
Maize	*Zea mays*	Grains
Chick pea	*Cicer arietinum*	Grains
Maize	*Zea mays*	Cobs
Walnut	*Juglans regia*	Shell
Almond	*Prunus amygdalus*	Chips
Barely	*Hordeum vulgare*	Grains

Spawn preparation

All the substrates were cleaned, washed and soaked in water and then boiled in plain water to soften them. The substrates were then cooled at room temperature and excess water was drained out by spreading them on wire mesh under shade followed by mixing with calcium carbonate and calcium sulphate in 1:4 ratio. The substrates were filled in cleaned bottles upto 3/4th of their capacity, plugged with non-absorbent cotton and sterilized in autoclave at 1.05 kg cm^{-2} pressure and 121 °C temperature for one and half hour.

Inoculation

The sterilized bottles containing different substrates were inoculated with master spawn under aseptic conditions and kept for incubation in B.O.D incubator at 25±2 ºC. These were arranged in completely randomised design with 4 replication for each substrate.

Observations recorded

The following observations were recorded

- Days taken for complete spawn run.
- Colour of spawn: colour of spawn was judged by visual observation.
- Texture (fluffy/cottony).
- pH:- pH of the spawn was measured with help of pH meter. 10 g of spawn was taken in a glass beaker with 25 ml of water. The contents in the beaker were stirred and pH was directly measured by dipping the pointed end of the instrument in the beaker containing spawn and water.

Results and Discussion

Spawn production on different substrates

The suitability of different substrates for the production of *Pleurotus florida* spawn was studied during 2020-2021 at Mushroom Technology and Training Centre, Division of Plant Pathology, SKUAST-Kashmir, FoA, Wadura. The experiment was laid out to study time required for complete spawn run, pH, colour and texture of spawn.

Time taken for complete spawn run

The data presented in Table 2 and Plate 1, revealed that complete spawn run of *Pleurotus florida* in seven different substrates *viz.*, walnut shells, almond chips, wheat grains, chickpea grains, maize cobs, maize grains and barley grains (check) varied significantly.

Minimum time was taken by walnut shells (6.75 days) and almond chips (7.00 days) for complete spawn run, barley grains (check) took (7.25 days) and maize grains (7.75 days) for complete spawn run and were statistically at par with each other. Time taken by wheat grains for complete spawn run was 8.25 days. All the other substrates took two to four days more than check for complete spawn run. Chickpea grains took 10.50 days for complete spawn run while maize cobs took maximum time of 12.25 days.

Table 2: Effect of different substrates on spawn run of *Pleurotus florida*

Substrate	Complete spawn run (days)*
Walnut shells	6.75
Almond chips	7.00
Chickpeas	10.50
Wheat grains	8.25
Maize grains	7.75
Maize cobs	12.25
Barley grains (check)	7.25
S.E (d)	0.23
C.D (p≤0.05)	0.49

*Data represents mean of 04 replications

Colour, texture and pH of spawn

The spawn prepared on different substrates showed significant difference in pH and it varied from 6.65 to 7.95 (Table 3). The pH 6.65 was recorded in walnut shell based spawn which was followed by almond chips pH 6.80 and these results were statistically at par. The pH 7.4 was recorded in wheat grains spawn which was at par with barley grains (check) spawn pH 7.4 and maize grains pH 7.4. The pH 7.39 was recorded in chickpea spawn while pH 7.9 was recorded in maize cob spawn. Spawn colour varied from brownish white in case of walnut shells and almond chips, light brown in chickpeas, whitish in case of grain substrates and maize cobs.

Texture of spawn varied from fluffy in walnut shells, almond shells, barley grains, wheat grains and maize grains to cottony in chickpeas.

Table 3: Effect of different substrates on colour, texture and pH of *Pleurotus florida* spawn

Substrate	Colour	Texture	Mean pH*
Walnut shells	Brownish-White	Fluffy	6.65
Almond chips	Brownish-White	Fluffy	6.80
Chick pea	Lightish brown	Cottony	7.37
Wheat grains	White	Fluffy	7.40
Maize grains	White	Fluffy	7.40
Maize cobs	White	Fluffy	7.95
Barley grains (check)	White	Fluffy	7.40
S.E (d)			0.15
C.D ($p \leq 0.05$)			0.33

*Data represents mean of 04 replications

All the substrates supported mycelial growth *of Pleurotus florida.* Fastest spawn run was recorded in walnut shells (6.75 days) and almond chips (7.00 days) followed by barley grains (7.25 days). In all the other substrates, spawn run was slower. These results are being reported for first time that walnut shells and almond shells took less time for complete spawn run of *Pleurotus florida*, the reason for the same could be the lignocellulolytic activity of *Pleurotus* species. Ligninolytic enzymes or ligninases are mainly comprised of laccases (Lac), lignin peroxidases (LiPs,), manganese peroxidases (MnPs), versatile peroxidases (VPs) and dye decolorizing peroxidases (DyPs).

a. Walnut shell chips b. Almond shell chips c. Chickpea grains

e. Barley Grains d. Wheat Grains c. Maize Grains

f. Maize cobs

Plate 1: Spawn run of *Pleurotus florida* on different evaluated substrates

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