Yield analysis of oyster mashroom (*Pleurotus ostreatus*) on *Ficus religiosa* leaves in combination with agricultural waste materials

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**Abstract**
Oyster mushrooms are one of the most common types of cultivated mushrooms in the world.

The current research was conducted to evaluate different agricultural wastes to improve the yield production of oyster mushroom. In the present research, five concentrations (T₁ = 100% Ficus leaves, T₂ = 75% Ficus leaves + 25% Agri. waste, T₃ = 50% Ficus leaves + 50% Agri. waste, T₄ = 25% Ficus leaves + 75% Agri. waste and T₅ = 0% Ficus leaves + 100% Agri. waste) of various agricultural wastes namely wheat straw, corn meal, cotton waste and thatch grass were used along with *F. religiosa* leaves. The experiment was conducted in under completely randomized design (CRD) with three replications. It was observed that the treatment T₃ expressed more yield (413.48 g) as compared to the other treatments viz., T₄ = 376.65g, T₅ = 340.91g, T₂ = 292.64g and T₁ = 250.49g, respectively, in all the three flushes. It was concluded that the yield production and biological efficacy of treatment 3 containing 50% Ficus leaves + 50% Agri. waste were highly useful to enhance the yield of oyster mushroom and can be used in future.

**Keywords:** *Ficus religiosa*; Growth chamber; Oyster mushroom; Yield assessment

**Introduction**
Almost 200 species of edible mushrooms are present in natural surroundings, but only 22 species are widely cultivated in different areas of the world. Oyster mushroom (*Pleurotus* species) is commercially grown on a large scale in many countries. It is an important commodity in the world mushroom market [1]. It belongs to phylum Basidiomycota, class Basidiomycetes and...
family Agaricaceae of the kingdom Fungi. Oyster mushroom is known as wood decomposers and grown on dung piles, water conduits, hilly ranges, in timberland, hardwood, on wood byproducts (paper, sawdust, pulp sludge), all cereal straws, sugarcane bagasse, corn cobs, coffee residues (stalks, coffee grounds, leaves, hulls), cottonseed hulls, soy pulp and banana fronds frequently during and after rain. The fruiting bodies of mushroom are pink, gray to dark brown in color with different sizes ranging from 4-15cm. The fruiting bodies of wild species appear in autumn; however, it is also observed during winter or in early warm springs. P. ostreatus is tolerant to low temperature but have high requirements of light because it is incapable to produce fruiting bodies under low light conditions [2].

Mushrooms are highly consumed due to their delicacy, aroma and texture. On dry weight basis, these contain about 39.91% carbohydrates, 17.54% proteins and 2.93% fats [3]. Some mushroom species are consumed due to excellent source of digestible proteins, fibers, carbohydrates and vitamins. Various edible species of Pleurotus are cultivated all over the world such as P. ostreatus, P. florida, P. salignus, P. columbinus, P. spodoleucus, P. pulmonarius and subspecies are P. sapidus, P. sajor-caju etc. Among all of these species, P. ostreatus is widely consumed and much easier to cultivate on biological waste materials due to its higher nutritional value [4].

P. ostreatus can be cultivated on several kinds of substrates/ waste material, having lignin and cellulose components, Due to its shorter growth period, less confronted by pest and disease as compared to the other consumable mushrooms, it can easily be managed [5]. Different agro-industrial wastes along with their different combinations have been used to evaluate the yield or quality production of P. florida and proved effective [6].

An experiment was conducted to analyze the nutritional values, minerals and growth of oyster mushroom by applying substrates in pure and mixed form. The pure substrate contained sawdust of Daniella olivery tree and the other substrate was a combination of (Mulainaner+ Daniella olivery tree) mixed with rice bran, rubber neck and calcium carbonate in different percentages, respectively. The growth limitations of mushroom picked from pure substrate was slightly healthier than the mixed substrate but the later one had the better nutritional compositions than the pure substrates [7].

F. religiosa is usually known as Bohar, in Subcontinent. It is a gigantic and semi-deciduous thinning out tree of 2100cm range. Its leaves are prevalent or elliptical with spikes like endings. It is one of the prehistoric tree in the God's creatures. It may be monoeccious or dioecious [8]. F. religiosa is a source of carbohydrates (cellulose, hemicelluloses, pectin) vitamin B6, copper, dietetic fibers, nitrogen, iron and magnesium. In therapeutic field, its leaves are used for Jaundice treatment, the sticky stuff of leaves or fruits may be used as therapy to treat snake bite. Its fruits are consumed by birds, livestock and other animals [9].

Agricultural waste material like sugarcane bagasse, paddy straw, maize cob and saw dust are valuable source for the production of oyster mushroom when they are mixed in various concentration, as they are rich source of cellulose therefor, they have a better capacity for the cultivation of mushroom [10].

Banana leaves having larger amount of cellulose in combination with wheat straw, rice straw and saw dust with different weight ratios are good compost for the production of Pleurotus species with an average daily temperature of 26-30°C and relative humidity of 80-100% [11].

Bioconversion of agriculture waste materials has been proved to be commercially helpful in the production of mushroom by using them as substrate [12].

In Pakistan, India and Bangladesh, the trade of oyster mushroom is increasing due to its pharmaceutical consumption and nutritious
inference. *F. religiosa* leaves are considerably used for the production of oyster mushroom along with other agricultural waste materials including corn meal straws of wheat, cotton waste and thatch grasses due to its promising source of essential nutrients. Ficus leaves enhance the production of mushroom and consequently prove to be helpful for minimizing the malnutrition [13]. Therefore, the present research was planned to use the *F. religiosa* leaves as agricultural waste substrate to increase the yield performance of *P. ostreatus*.

**Aim of the experiment**

Nearly 75% population of Pakistan indirectly depends on agriculture and most of the people are predominately attached to the farming community. The present study was designed to assess the suitability of spawn in production and also to cultivate the *P. ostreatus* mushroom on different nutrient rich agricultural waste materials.

**Materials and Methods**

The experiment was conducted, during 2017-18, in the mushroom cultivation chamber, Institute of Horticulture sciences, UAF under CRD with three replications. In the current research, *F. religiosa* leaves were used in combination with different agricultural waste materials viz., wheat straw, corn meal, waste of cotton and thatch grass by using five treatments with three replications (Table 1). Gypsum and wheat bran were added to maintain the pH level of the substrate.

**Table 1. Formation of treatments according to their proportion**

| Treatments | Substrate Ratios % |
|------------|---------------------|
|            | *Ficus religiosa* leaves | Agricultural waste material (wheat straw+ cotton waste+ corn meal+ thatch grass) |
| T1         | 100%                 | 0%                       |
| T2         | 75%                  | 25%                      |
| T3         | 50%                  | 50%                      |
| T4         | 25%                  | 75%                      |
| T5         | 0%                   | 100%                     |

**Substrate preparation**

The leaves of *F. religiosa* were used along with other additive substrates namely wheat straw, corn meal, waste of cotton and thatch grass for the cultivation of oyster mushroom. All substrates were collected from the fields of Department of Forestry and Agronomy, UAF. The leaves were chopped into 5-7cm pieces, soaked in fresh water for 6-8hrs and sterilized in hot water for 2-3hrs at Mycology Lab, Department of Plant Pathology, UAF. The substrates after sterilization were spread on polyethylene sheet under sunlight for 10 days to transpire the excessive water.

Substrates such as wheat straw, cotton waste, corn meal and thatch grass were chopped into 5-7cm pieces and soaked in fresh water for 6-8hrs and sterilized in hot water for 2-3hrs. After that, the substrate was dried till it contains 60-70% moisture contents. Spawning was done at 4% by wet weight of the prepared substrate. Cultivation was done in polyethylene bags of 0.002cm thickness [14].

**Cultivation of oyster mushroom**

The fermented substrates were packed in heat tolerant polypropylene bags (24.8×17cm). The bags contained 500g of substrates along with different concentrations of treatments. Afterwards, the bags were air tightened by using a rubber band and autoclaved at 121°C with 15-20psi pressure for 20min. Subsequently, 15g fresh spawn culture was inoculated in each bag under controlled conditions. The temperature and humidity of the growth chamber were maintained at 20-25°C and 80-90%, correspondingly. The fruiting bodies were sprouted after 7-10 days.
Calculation of biological efficiency
Biological efficiency of each bag of the substrate was calculated by using the formula [15].

\[
\text{Biological Efficiency (\%) = \frac{\text{Fresh weight of mushroom harvested}}{\text{Substrate dry matter}} \times 100}
\]

Statistical analysis
The statistical analysis was performed through statistical software Statistix v. 8.1. The means of treatments were compared using Fisher’s least significant difference (LSD) test at 0.05% probability level.

Results and Discussion
All the substrates supported the production of *P. ostreatus*. However, the time required for the initiation and completion of fruiting bodies was significantly reduced by T\(_3\) having 50% of *F. religiosa* leaves + 50% of agri. waste material compared to - T\(_4\), T\(_5\), T\(_2\) and T\(_1\), respectively.

This experiment was conducted to find out the suitability of locally available substrates for the production of *P. ostreatus*. *F. religiosa* leaves along with different agriculture waste materials (cotton waste, wheat straw, corn meal and thatch grass) were applied and the significant results were obtained. In general, *F. religiosa* leaves along with other agri. waste material (50:50) were found to be superior to the other treatments made during this study.

It was examined that among all the treatments T\(_3\) (50% Ficus leaves + 50% agri. waste) took minimum days (26) for the development of 100% mycelial growth in bags. On the other hand, the longest period was taken by the treatment T\(_1\) (100% Ficus leaves) 67 days for the formation of pinheads. The results of the other treatments for the formation of pinheads were; T\(_4\) (25% Ficus leaves + 75% agri. waste) took 57 days, treatment T\(_5\) (0% Ficus leaves + 100% agri. waste) took 60 days and treatment T\(_2\) (75% Ficus leaves + 25% agri. waste) took 62 days, respectively.

It was observed that the treatment T\(_3\) (50% Ficus leaves + 50% agri. waste) showed statistically better results than the other treatments [17, 18].

After the complete development of pinheads, the fruiting bodies started to emerge from the bags. It was examined that among all the treatments T\(_3\) (50% Ficus leaves + 50% agri. waste) started to produce the fruiting bodies within nine days while the treatment T\(_1\) (100% Ficus leaves) took 21 days. The remaining treatments took different no. of days to produce fruiting bodies, T\(_4\) (25% Ficus leaves + 75% agri. waste) 12 days, T\(_5\) (0% Ficus leaves + 100% agri. waste) 16 days and T\(_2\) (75% Ficus leaves + 25% agri. waste) 19 days, respectively. Overall, the treatment T\(_3\) (50% Ficus leaves + 50% agri. waste) showed highly significant results than the other treatments. The same outcome was given by [19] while using waste paper in supplementation with rice bran.
In view of the above study, it can be concluded that the yield contributed characteristics and biological efficiency of

Table 2. Assessment for the growth development of oyster mushroom (Days)

| Treatments          | 100 % Mycelial growth | Pin head formation | Dev. of fruiting bodies |
|---------------------|-----------------------|-------------------|-------------------------|
| T₁                  | 35.40 ± 0.011 A       | 67.40 ± 1.02 A    | 21.20 ± 0.58 A          |
| T₂                  | 31.60 ± 0.007 B       | 62.00 ± 1.09 B    | 19.20 ± 0.50 B          |
| T₃                  | 26.81 ± 0.010 E       | 54.40 ± 1.02 D    | 9.00 ± 0.70 E           |
| T₄                  | 28.82 ± 0.014 D       | 57.60 ± 1.07 C    | 12.60 ± 0.50 D          |
| T₅                  | 30.21 ± 0.013 C       | 60.80 ± 1.11 B    | 16.20 ± 0.58 C          |

LSD of treatment means were compared at significance at p ≤ 0.05, T₁ = Ficus leaves 100%, T₂ = Ficus leaves 75% + agri. waste 25%, T₃ = Ficus leaves 50% + agri. waste 50%, T₄ = Ficus leaves 25% + agri. waste 75%, T₅ = Ficus leaves 0% + agri. waste 100%

Yield analysis
The average weight of mushroom yield in first flush was higher in T₃ (150.75g) while lowest in treatment T₁ 94.31g, while the remaining treatments namely T₄, T₅ and T₂ had 137.33g, 123.84g and 108.41g yield of mushroom, respectively, in the first flush (Table 3). The results of the present experimental trial regarding yield in first flush are in close conformity with the investigation of [20] whose results showed 89-146g yield of mushroom from the first flush.

Similarly, T₃ produced average yield of 136g in the second flush of mushroom as compared to other treatments which include viz., T₁ (81.21g), T₄ (122.24g), T₅ (118.48g) and T₃ (98.314g), respectively (Table 3). The third and last flush of mushroom expressed less amount of yield with low nutrient values. The high weight of yield was shown by T₃ treatment (126.15g) whereas T₁ treatment expressed least amount of mushroom that was 74.96g. Likewise, all other treatments such as T₄, T₅ and T₂ exhibited 117.08g, 98.90g and 85.90g of mushroom yield, respectively (Table 3). The total mean yield observed from all the treatments of this experiment varied from 413-250g and are considerably in line with the findings of [17] who used the wheat straw and [21] they used 50% sisal leaves + 50% sisal boles and observed 450g of maximum mushroom yield.

It was observed that the biological efficiency of *P. ostreatus* mushroom varied to utilize different substrates for producing yield. Highest biological efficiency 82.70% recorded in case of T₃ (50% Ficus leaves+ 50% Agri. waste) followed by T₄ (25% Ficus Leaves+ 75% Agri. waste) 75.32%, T₅ (50% Ficus Leaves+ 100% Agri. waste) 68.18%, T₂ (75% Ficus Leaves+ 25% Agri. Waste) 58.52% and Treatment T₁ (100% Ficus Leaves) 50.09% respectively (Table 3). The outcomes of this experiment are in line with [22, 23] who reported 85% and 77% of biological efficiency of *Pleurotus* spp. in paddy straw and wheat straw.

Table 3. The yield and biological efficiency of oyster mushroom on different treatments

| Treatments          | 1st flush | 2nd flush | 3rd flush | Total yield | B.E (%) |
|---------------------|-----------|-----------|-----------|-------------|---------|
| T₁                  | 94.31 ± 1.22 E | 81.21 ± 0.96 E | 74.96 ± 1.19 E | 250.49 ± 2.67 E | 50.09 ± 0.53 E |
| T₂                  | 108.41 ± 1.22 D | 98.31 ± 1.29 D | 85.90 ± 0.93 D | 292.64 ± 2.39 D | 58.52 ± 0.47 D |
| T₃                  | 150.75 ± 0.97 A | 136.58 ± 0.92 A | 126.15 ± 0.87 A | 343.98 ± 1.42 A | 82.70 ± 0.28 A |
| T₄                  | 137.33 ± 1.04 B | 122.24 ± 0.99 B | 117.08 ± 0.99 B | 376.65 ± 1.16 B | 75.32 ± 0.23 B |
| T₅                  | 123.84 ± 1.02 C | 118.48 ± 0.92 C | 98.90 ± 1.10 C | 340.91 ± 0.95 C | 68.18 ± 0.19 C |

LSD of treatment means were compared at significance at p ≤ 0.05, T₁ = Ficus leaves 100%, T₂ = Ficus leaves 75% + agri. waste 25%, T₃ = Ficus leaves 50% + agri. waste 50%, T₄ = Ficus leaves 25% + agri. waste 75%, T₅ = Ficus leaves 0% + agri. waste 100%, B.E= Biological efficiency

Conclusion
In view of the above study, it can be concluded that the yield contributed characteristics and biological efficiency of T₃ (*F. religiosa* leaves along with other agri. waste material (50:50) was maximum 413.48g in minimum days. The combination of main substrate along with
other agri. waste materials for the production of oyster mushroom showed an opportunity for commercial cultivation when they were used in combined form.

Authors’ contributions
Conceived and designed the experiments: NA Khan, Performed the experiments: A Tahir, Analyzed the data: K Ikram & M Ashfaq, Contributed materials/ analysis/tools: HM Aatif & Ch. MS Hanif, Wrote the paper: MZ Mansha & HMU Aslam.

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