

Influence of addition of different cereal and leguminous flours on spawn growth of *Pleurotus ostreatus*

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ABSTRACT

Oyster mushroom is one of the most widely cultivated mushrooms in the world. Spawn production is a crucial part of the mushroom growing process. Experiments were conducted to determine the effect of addition of different cereal and leguminous flour (rice, wheat, maize, gram, chickpea, lentil, soybean and black gram) at different concentrations (2, 4 and 6%) on spawn growth of *Pleurotus ostreatus*. Among cereal flours, the maximum Mycelial Run Rate (MRR) was observed at 6% wheat flour (10.95 mm day⁻¹) as compared to other treatments followed by 2% rice flour (10.47 mm day⁻¹). In case of leguminous flours, the MRR at 2% gram and 4% black gram flour (9.26 mm day⁻¹) were the maximum and statistically on par with each other followed by 2% soybean flour (8.33 mm day⁻¹). Out of all tested additives, 6% wheat flour gave the maximum MRR as well as took lesser time to complete mycelial growth as compared to other additives. The least MRR was observed in control (6.96 mm day⁻¹) and took 15th days. The results indicated that addition of supplements enhances the spawn growth and minimizes the days of spawn preparation.

Keywords: *Pleurotus ostreatus*, spawn, Mycelial Run Rate, cereal and leguminous flour

Mushrooms can be produced on a variety of substrates and are consumed by people of all ages (Nongthombam *et al.*, 2021) due to its rich nutritional values like proteins, minerals, vitamins like vitamin B complex, C and D (Ahmed *et al.*, 2009). Fresh mushrooms contain 85-90% moisture, 3% protein, 4% carbohydrate, 0.3-0.4% lipids and 1% vitamins and minerals (Miah *et al.*, 2017). Oyster mushroom (*Pleurotus* species) has a wide range of uses in both food and medicine, and are a strong source of protein, vitamins, and minerals (Etich *et al.*, 2013). In addition, they are reported to have antibacterial, antitumor, antithrombotic, hypoglycemic, anti-inflammatory, anti-hypertensive and other properties (Gunde, 1999). Cultivation of oyster mushroom is becoming popular

in developing countries because of easy and cost-effective cultivation technology (Sud and Jandaik, 2007). It occupies third place in commercially cultivated mushroom in the world (Patel *et al.*, 2012). Out of 40 reported different species (Sud and Jandaik, 2007), *Pleurotus ostreatus* is the second most popular species in the world market (Sanchez *et al.*, 2010).

Spawn act as the starting point in mushroom production (Royse, 2003; Stanley and Awi-Waadu 2010) and regulates the productivity of the mushroom. There are reports indicating that for vigorous growth of mushroom mycelium is significantly impacted by the various additives (Shah *et al.*, 2004).

Keeping in view the importance and potential of growing oyster mushrooms in both rural and urban areas of the country, the experiment was conducted with the aim to study the influence of different organic additives of cereal and leguminous flour on spawn growth of *Pleurotus ostreatus*.

MATERIALS AND METHODS

Experimental site

The investigations were carried out in 2021–2022, in Mushroom Laboratory, Department of Plant Pathology, CSKHPKV, Palampur, HP (India).

Pure culture and spawn production

Pure culture of oyster mushrooms (*Pleurotus ostreatus*) was obtained from the ICAR- Directorate of Mushroom Research, Solan and maintained on Potato Dextrose Agar (PDA) slants. Mushroom spawn was prepared on wheat grains substrate as per standard procedure (Sinden, 1932).

Mushroom spawns were supplemented with cereal grain and leguminous flours at different concentration i.e. 2, 4 and 6%. Wheat grains were used as substrate for spawn production. Grains and additives were sterilized separately in bottles in an autoclave at 22 lb psi for 2 hours. The bottles and additives were well shaken to avoid clumping of grains. Under aseptic conditions, organic additives were added to grains at different concentrations (2, 4 and 6%) and then inoculated with the pure culture. The inoculated spawn bottles were incubated at 24±1 °C without shaking and observations were made on 3rd, 5th, 7th, 9th, 11th and 13th days or until complete mycelial growth was observed. The growth of the spawn was measured in terms of Mycelial Run Rate (MRR) (mm day⁻¹).

Mycelial run rate (MRR)

Mycelial run rate was measured and calculated as per Tarushi *et al.*, (2020).

Statistical analysis

The experiment was set in Complete Randomized Design (CRD) and the data were subjected to Analysis of variance (ANOVA). Critical difference (CD) were calculated at five percent level of significance for comparison with other treatment (Steel *et al.*, 1997).

RESULT AND DISCUSSION

Various organic additives were used in three different concentrations i.e. 2, 4 and 6% and mixed in wheat grain to study the days required for full mycelial colonization of grain spawn and to calculate the rate mycelial growth per day. The data obtained on colonization of grains on addition of cereal and leguminous flours, and the number of days needed for spawn preparation is presented in Table 1. In the spawn development of oyster mushroom significant variation was observed on addition of various cereal flours at different concentration. Addition of 6% wheat flour took the least time i.e.11 days followed by 2% rice flour, which took 13 days for complete spawn development in glass bottles. Significant variation on

Table 1. Days required for complete mycelial growth of *Pleurotus ostreatus* at different concentration of cereal and leguminous flour

| Organic additives | Days taken to complete spawn colonization | | |
|-------------------|---|----|----|
| | 2% | 4% | 6% |
| Wheat flour | 13 | 13 | 11 |
| Rice flour | 13 | 13 | 13 |
| Maize flour | 15 | 15 | 15 |
| Gram flour | 13 | 15 | 15 |
| Soybean flour | 15 | 15 | 15 |
| Black gram flour | 15 | 13 | 15 |
| Lentil flour | 15 | 15 | 15 |
| Chick pea flour | 15 | 15 | 15 |
| Control | 17 | | |

complete colonization of grains was observed on various leguminous flours at different concentrations. In case of leguminous flours 2% gram and 4% black gram flour required minimum time (13 days) to complete mycelial growth. On control, the maximum period (17 days) for complete mycelial growth was observed. Thus, overall it was observed that addition of 6% wheat flour supported early growth of mycelium in spawn preparation.

The highest MRR was observed on addition of 6% wheat flour (11.48 mm day⁻¹) followed by 4% wheat flour (10.89 mm day⁻¹) and 2% rice flour (10.60

mm day⁻¹) and lowest MRR was observed in control (6.96 mm day⁻¹) (Table 2). The mycelial run rate on 6%, 4% and 2% wheat flour additive were significantly different with each other whereas there was no significant difference with respect to the mycelial run rate on 2 and 4% concentration of rice flour. With respect to leguminous flours, highest MRR was observed on 2% gram and 4% black gram flour (9.26 mm day⁻¹) and were statistically on par while lowest MRR was observed on control (6.96 mm day⁻¹) (Table 2). The percentage increase in the mycelial growth of oyster mushroom mycelial growth was observed maximum in 2% gram and 4% black gram flour.

Table 2. Mycelial Run rate of *Pleurotus ostreatus* at different concentrations of cereal flour

| Organic additives | Conc | Mycelial run rate (mm/day) | | | | | | |
|-------------------|------|----------------------------|-------|-------|-------|--------|--------|--------|
| | | 3 DAI | 5 DAI | 7 DAI | 9 DAI | 11 DAI | 13 DAI | 15 DAI |
| Wheat flour | 2% | 4.95 | 6.97 | 7.43 | 10.56 | 9.58 | 9.26 | 8.02 |
| | 4% | 6.63 | 7.02 | 7.85 | 10.89 | 9.86 | 9.27 | 8.04 |
| | 6% | 8.39 | 8.40 | 9.42 | 11.48 | 10.95 | 9.26 | 8.03 |
| Rice flour | 2% | 6.79 | 8.47 | 9.41 | 10.60 | 10.47 | 9.29 | 8.06 |
| | 4% | 6.82 | 7.05 | 8.33 | 8.96 | 10.05 | 9.27 | 8.04 |
| | 6% | 5.22 | 5.75 | 6.95 | 7.82 | 9.12 | 9.27 | 8.03 |
| Maize flour | 2% | 5.22 | 5.09 | 6.46 | 7.15 | 7.50 | 8.04 | 8.01 |
| | 4% | 6.79 | 7.72 | 7.24 | 8.07 | 8.59 | 8.36 | 8.03 |
| | 6% | 6.08 | 7.15 | 6.63 | 7.31 | 7.76 | 8.10 | 8.03 |
| Gram flour | 2% | 6.68 | 7.99 | 9.29 | 8.95 | 8.57 | 9.26 | 8.03 |
| | 4% | 6.72 | 7.00 | 8.58 | 9.43 | 9.14 | 8.07 | 8.02 |
| | 6% | 5.00 | 4.40 | 5.43 | 6.45 | 6.60 | 7.55 | 7.37 |
| Soybean flour | 2% | 6.89 | 8.07 | 7.90 | 8.71 | 8.93 | 8.33 | 8.01 |
| | 4% | 4.07 | 6.09 | 5.35 | 5.86 | 6.43 | 7.13 | 7.03 |
| | 6% | 5.22 | 6.13 | 6.57 | 6.96 | 7.48 | 7.73 | 8.03 |
| Black gram flour | 2% | 5.12 | 7.79 | 7.18 | 8.06 | 8.41 | 8.07 | 8.02 |
| | 4% | 6.82 | 8.10 | 8.65 | 9.39 | 9.16 | 9.26 | 8.03 |
| | 6% | 5.28 | 5.71 | 6.04 | 6.47 | 6.99 | 7.56 | 7.37 |
| Lentil flour | 2% | 3.77 | 5.08 | 5.47 | 5.82 | 6.40 | 7.36 | 7.22 |
| | 4% | 5.11 | 7.11 | 6.08 | 7.25 | 7.51 | 7.74 | 8.02 |
| | 6% | 5.13 | 6.05 | 5.75 | 8.04 | 8.16 | 8.10 | 8.04 |
| Chick pea flour | 2% | 3.77 | 5.10 | 5.75 | 6.15 | 6.70 | 7.58 | 7.68 |
| | 4% | 5.18 | 7.13 | 7.20 | 7.84 | 8.22 | 8.15 | 8.03 |
| | 6% | 5.45 | 7.34 | 6.56 | 7.70 | 7.66 | 7.88 | 8.05 |
| Control | | 3.41 | 4.12 | 5.04 | 5.56 | 5.68 | 6.59 | 6.96 |
| CD (5%) | | 0.23 | 0.34 | 0.65 | 0.49 | 0.34 | 0.33 | 0.36 |

DAI = Days after inoculation

Singh *et al.* (2017) in his studies evaluated the influence of different organic additives (pulses flour) on spawn production of oyster mushroom (*Pleurotus djamor*) and found maximum mycelial growth (100.00 mm) in 2% pigeon pea flour and minimum in control (20 days). Singh *et al.* (2021), reported maximum growth of *Cordyceps militaris* mycelium in maize flour while pigeon pea flour was reported best organic additive for spawn production. Kumar *et al.* (2019) studied the effect of different inorganic and organic additives on spawn growth of two strains (CI-17-04 and CI-17-08) of milky mushroom (*Calocybe indica*) and found the maximum mycelial growth (90.00 mm) in rice powder @ 1%. A healthy and vigorous spawn is a prerequisite to get healthy fruit bodies of mushroom. Different grains and different additives have been tried in the past to get the faster spawn growth in the production of oyster mushrooms. With respect to mycelial run rate (MRR) also, addition of 6% wheat flour supported faster growth of mycelium during spawn preparation.

CONCLUSION

Spawn is an important component for mushroom cultivation that influences mushroom yield. The study concluded that both cereal and leguminous additives enhanced the spawn growth, which was indicated by lesser number of days and more MRR during spawn production as compared to control. However, it was observed that the 6% wheat flour best supported spawn growth of the oyster mushroom followed by 2% rice flour amongst cereal additives. In case of leguminous flour, 2% gram and 4% black gram best supported the spawn growth followed by 2% soybean flour. Among both cereal and leguminous additives, the 6% wheat flour was found to be the best additive for spawn production of oyster mushroom.

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