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Valorization Of Agro-Residues For Sustainable Cultivation Of *Pleurotus* Spp.: A Circular Bioeconomy Approach To Mushroom Biotechnology

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ABSTRACT

Mushroom cultivation presents considerable nutritional, ecological, and economic advantages, especially in developing countries. However, poor management of agricultural waste raises significant environmental concerns and threatens food security. This study aimed to explore the optimal substrate selection for cultivating *Pleurotus* spp. The experimental design utilized a completely randomized design (CRD) methodology, incorporating three replicates for each substrate and mushroom species combination. The data collected underwent a thorough descriptive statistical analysis, emphasizing yield comparison and colonization efficiency. The highest yield was observed with rice straw (760 g/kg) for *P. ostreatus*, followed by wheat straw (780 g) for *P. florida* and soybean straw (600 g) for *P. djamor*. This species exhibited clear substrate preferences, with soybean straw consistently identified as one of the most effective options, yielding high outputs and biological efficiency (BE) across various studies. These findings underscore the critical role of substrate selection in optimizing the cultivation of Pleurotus mushrooms.

Keywords: Pleurotus spp.; Mushroom biotechnology; Biological efficiency; Lignocellulosic substrates; Agricultural residues.

INTRODUCTION

Mushroom cultivation offers significant nutritional, ecological, and economic benefits, particularly in developing countries. Mushrooms are rich in proteins, carbohydrates, vitamins, and minerals, making them an excellent supplement to cereal-based diets. They also possess medicinal properties due to bioactive compounds like polysaccharides and triterpenoids (Singh, 2023; Verma, 2024; Losoya-Sifuentes *et al.,* 2025; Thakur and Singh, 2013). Mushroom production can alleviate food insecurity, create job opportunities, and contribute to economic growth (Thakur, 2020). The industry utilizes agroforestry waste, aligning with sustainable waste management practices (Chiu *et al*., 2000). Despite its potential, the sector faces challenges such as inadequate expertise and a lack of supportive policies in developing nations (Itubochi *et al*., 2025). Innovations in cultivation techniques, processing equipment, and value-added products are enhancing the industry's efficiency and marketability (Singhal *et al.,* 2019). Conservation of mushroom biodiversity is crucial for genetic resources and breeding programs to meet diverse consumer demands (Chiu *et al*., 2000).

Agricultural waste mismanagement poses significant environmental and food security challenges. Large quantities of agro-waste are generated globally, with improper disposal leading to pollution and resource loss (Sabiiti, 2011). However, these wastes can be valuable resources if properly managed. Strategies for sustainable agro-waste management include reduction, reuse, and recycling (Koul *et al.,* 2021). Urban agro-producers employ various management practices such as waste reduction, utilization, segregation, and composting (Karani *et al.,* 2021). Agro-wastes can be used to enhance soil fertility, improve crop yields, and increase livestock productivity, thereby contributing to food security (Sabiiti, 2011). Additionally, agro-industrial wastes can be valorized into value-added products like biofuels, biofertilizers, and food ingredients. Proper management and utilization of agro-wastes can help address food insecurity, reduce environmental pollution, and contribute to sustainable agriculture and circular bioeconomy (Sabiiti, 2011; Koul *et al*., 2021; Mohanta *et al.,* 2025).

Agricultural waste management in India presents both challenges and opportunities. Large amounts of lignocellulosic waste from agriculture and food processing industries pose environmental concerns if disposed of improperly (Ravindran and Jaiswal, 2015; Kumar *et al*., 2022). However, this waste can be effectively utilized for mushroom cultivation, offering an eco-friendly solution to waste management while producing nutritious food (Khan *et al.* 2021; Adegbeye *et al.* 2020). Various mushroom species, including Pleurotus, can colonize and degrade a wide range of agricultural wastes such as rice husks, corn cobs, and cotton waste (Rakib *et al.* 2020; Barshteyn & Krupodorova, 2016). Mushroom cultivation not only reduces waste but also produces valuable enzymes and fruiting bodies (Kumla *et al.* 2020). Additionally, recent research explores the potential of using agricultural waste and mycelium to develop sustainable construction materials, further expanding the applications of waste utilization (Voutetaki and Mpalaskas, 2024). These approaches offer promising solutions to India's agricultural waste management challenges.

MATERIALS AND METHODS

The experimental cultivation of *Pleurotus ostreatus*, *Pleurotus florida*, and *Pleurotus djamor* was carried out under controlled laboratory conditions at the Department of Botany, Dr. Babasaheb Ambedkar Marathwada University in Chhatrapati Sambhajinagar, India. Throughout the study, a dedicated dark incubation room was maintained, with humidity levels regulated between 70–90% and temperatures kept at 24–28°C, ensuring precise monitoring through the use of a thermo-hygrometer.

Substrate Selection and Preparation: Six commonly available agricultural waste materials, viz rice straw, wheat straw, soybean straw, cotton plant waste, pigeon pea straw, and maize stalks, were selected based on their regional availability and lignocellulosic composition. The substrates were chopped into lengths of 3 to 5 cm and thoroughly washed in clean water to eliminate any dirt and impurities. They then underwent chemical sterilization using a combination of 0.5% formaldehyde and 0.1% fungicide (Bavistin) to prevent microbial contamination. After soaking for 12 hours, the substrates were drained to remove excess water, ensuring an optimal moisture content of approximately 65%, as confirmed by the tactile compression method (Sonneveld & Voogt, 2009).

Spawn Preparation and Inoculation: Pure spawn cultures of *P. ostreatus*, *P. florida*, and *P. djamor* were obtained from certified laboratories. Each sterilized substrate was inoculated with 5% (w/w) grain spawn under aseptic conditions. The inoculated substrates were then placed into perforated polypropylene bags (30 x 45 cm) to ensure adequate aeration and gas exchange. Each bag was securely sealed and incubated in a dark chamber (Thakur and Rathod, 2021).

Incubation and Mycelial Monitoring: The incubation period lasted for 12 days, during which the growth of the mycelium was monitored at intervals of the 3rd, 6th, 9th, and 12th days. The progress of growth was documented based on surface colonization, where "++++" signified full colonization. Mycelial colonization was assessed visually and scored using a system calibrated according to previous studies.

Fruiting and Harvesting: After complete mycelial colonization, the bags were moved to fruiting conditions, which included 8 to 10 hours of light exposure each day and maintaining a relative humidity of 85 to 90% using hand water sprayers. Daily monitoring of temperature and moisture levels was conducted. Fruiting bodies were harvested once they matured, indicated by cap flattening and the onset of spore release, and the total yield was recorded in grams per kilogram of dry substrate using a calibrated digital weighing scale (Thuy and Suzuki, 2019).

Experimental Design and Replication: The experimental design employed a completely randomized design (CRD) methodology, incorporating three replicates for each combination of substrate and mushroom species. The data obtained were subjected to a comprehensive descriptive statistical analysis, focusing on yield comparison and colonization efficiency.

RESULTS AND DISCUSSION

The colonization of mycelium was visually discernible by the twelfth day of incubation. The substrate-specific colonization dynamics for *Pleurotus ostreatus, P. florida*, and *P. djamor* are summarized in Table 1. For *Pleurotus ostreatus*, the highest yield was recorded with rice straw (760 g/kg), followed by moderate yields from wheat (700 g), soybeans (660 g), and pigeon peas (540 g). In contrast, the lowest yield was observed with maize straw (340 g/kg). This species demonstrated a clear preference for rice straw, likely attributable to its optimal porosity and favorable cellulose-to-lignin ratio, which enhances enzymatic degradation. Numerous studies have explored the growth and yield of *Pleurotus ostreatus* on various lignocellulosic substrates, with rice straw consistently emerging as one of the most effective options, yielding high outputs and biological efficiency (BE) across multiple investigations (Nasreen *et al*., 2016; Obodai *et al*., 2003; Biswas *et al*., 2023). Wheat straw also performed well, generally ranking second in yield. Additionally, soybean straw exhibited potential in one study, surpassing wheat straw (Patil, 2024).

In the case of *Pleurotus florida*, the highest yield was recorded with wheat straw (780 g/kg), with moderate yields from soybean (620 g), cotton (560 g), and rice straw (580 g). The lowest yield came from maize straw (320 g/kg). Here, *P. florida* thrived on wheat straw, a substrate known for facilitating higher protein-bound polysaccharides. Maize was the least efficient substrate, likely due to its high lignin content and dense cell walls. Similar studies have shown that wheat straw consistently emerged as an excellent substrate, yielding high mushroom production and biological efficiency for *P. florida*. Notably, paddy straw and cowpea pod residues also demonstrated high yields for *P. florida* (Iqbal *et al.*, 2016; Hiwale, 2021; Sufyan *et al.*, 2021).

For *Pleurotus djamor*, the highest yield was achieved with soybean straw (600 g/kg), followed by moderate yields from cotton (520 g) and maize (500 g). The lowest yield was recorded for rice and pigeon pea straw (200 g/kg each). This species displayed distinct substrate preferences, with soybean straw proving to be the most effective, possibly due to its rich nitrogen content and soft fiber composition. The low yield on rice straw, despite complete colonization, suggests potential issues related to substrate texture or nutritional imbalances. Recent studies have consistently shown that soybean straw is an excellent substrate for cultivating various Pleurotus species. For *P. djamor,* soybean straw yielded the highest biological efficiency (BE) of 109.4% (Deshmukh & Deshmukh, 2016). Rice straw combined with cocopeat and rice bran (7:3:1 ratio) proved effective for *P. djamor,* producing the highest yield (256.6 g) and BE (31.10%) (Zurbano et al., 2017). These findings highlight the importance of substrate selection in optimizing Pleurotus mushroom cultivation.

Table 1: Mycelial Growth and Yield Analysis of *Pleurotus* spp. on Different Agro-Wastes

Sr. No.	Substrate	Mycelial Colonization (12 Days)	Yield of *P. ostreatus* (g/kg)	Yield of *P. florida* (g/kg)	Yield of *P. djamor* (g/kg)
1	Rice Straw	++++	**760 ± 20**	580 ± 18	200 ± 15
2	Wheat Straw	++++	700 ± 25	**780 ± 22**	340 ± 16
3	Soybean Straw	++++	660 ± 22	620 ± 19	**600 ± 21**
4	Cotton Plant Waste	++++	500 ± 15	560 ± 17	520 ± 20
5	Pigeon Pea Straw	++++	540 ± 14	420 ± 13	200 ± 12
6	Maize Straw	++++	340 ± 12	320 ± 11	500 ± 18

"++++" = 100% colonization; ± = Standard deviation (triplicate values averaged).

Fig 1 Experimental setup for cultivation of *P. ostreatus*, *P. florida*, and *P. djamor*

CONCLUSION

This study highlights the significant effect of substrate selection on the yield performance of *Pleurotus* spp. during controlled incubation. Rice straw achieved the highest yield for *P. ostreatus*, while wheat and soybean straw were optimal for *P. florida* and *P. djamor*, respectively. Despite uniform mycelial colonization, yield variability emphasizes the importance of substrate-specific biochemical composition and structural characteristics. These findings align with previous research on lignocellulosic decomposition efficiency and advocate for strategic substrate use in scalable, sustainable mushroom cultivation within agro-waste valorization frameworks.

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