Global Research journal of Natural Science and Technology (Grjnst)
Volume: 03 - Issue 3 (2025)
p.ISSN: 2790-7643 e.ISSN: 2790-7651

Optimizing Oyster Mushroom (Pleurotus djamor) Yield, Effects of Spawn Rates on Cotton and Rice Cellulose Waste Substrates

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Abstract: *The study investigated the influence of varying spawn rates on the growth performance and yield of Pleurotus djamor (pink oyster mushroom) cultivated on cotton waste and rice cellulose waste substrates. Spawn rates of 15, 25, 35, 45, and 55 g per 500 g dry substrate were evaluated under a completely randomized design with four replications. Growth parameters, including time to pinhead formation, fruiting body maturation, number of flushes, bunches, and fruiting bodies per bunch, were recorded, while yield parameters such as fresh and dry yield, total soluble solids, protein content, and biological efficiency were assessed post-harvest. Results revealed that increasing spawn rate significantly enhanced growth and yield attributes. The 55 g spawn rate on cotton waste substrate achieved the best performance, producing the earliest pinhead formation (33 days), fastest fruiting body maturation (5.01 days), and the highest fresh yield (41.62%). However, maximum biological efficiency (56.63%) was obtained at the 25 g spawn rate. Overall, cotton waste proved to be a superior substrate compared to rice cellulose, supporting faster growth and higher yield. These findings highlight that optimizing spawn rate and substrate selection can significantly improve Pleurotus djamor production efficiency, providing a sustainable strategy for recycling agro-wastes into valuable edible biomass.*

Keywords: *Pleurotus djamor, spawn rate, cotton waste, rice cellulose, mushroom yield, biological efficiency, sustainable cultivation*

1. Introduction

Mushrooms have been described as enormous scale life forms with specific natural bodies, which are recognizable to the naked eye. Mushrooms have a spot with the class Basidiomycetes (Pelczar*et al.*, 2003). Study of mushrooms is called mycology, got from old Greek word "mykes". Pakistan being an agricultural country having 65 percent of its total population in this sector, does farming generally in an indirect way. Agriculture is the biggest section of the country that is responsible for giving employment opportunities to at least 70% of total work power (Mueller and Gawley, 1983; Jan et al., 2025). Growing oyster mushroom is getting progressively mainstream worldwide due to their ability to develop at a range of temperatures using different lignocelluloses (Khan and Garcha, 1984; Pidgeon and Anderson, 1981). Oyster mushrooms live wildly on parts of plants which are commonly poor in nutrients. For development of mushroom, lignocellulose materials are sufficient, for example wood shavings, corn cobs, every single grain straw, paper, sawdust, nutshells and vegetable materials and food industrial wastes. (Yildiz and Ertekin, 1996; Jan et al., 2025).

The mushroom development is a lucrative business and *Pleurotus* species are palatable mushroom having good nutritional qualities and flavor. It is native to the backwoods of uneven territories and is cultivated in subtropical regions. The innovation of fake development of *Pleurotus* species is to some degree late advancement, consolidation of nontraditional harvests in existing horticultural framework can help in improving the social just as monetary status of little ranchers. *Pleurotus* species have around 80-90% water, 4% protein, 4.5% sugars, 0.2% fats, 1.5% minerals and other nutrients (Tewari, 1986). As among the distinctive developed edible fungi, species of *Pleurotus* are the most straightforward and least expensive to develop and are preferable in customer perspectives over the by and large developed catch mushroom. *Pleurotus florida* forces cell reinforcement, immune stimulant, anti-tumor and calming exercises. The normal preferred position of mushroom development in India is because of the nearness of occasional variety, wealth and accessibility of assortment of agro-squanders at low costs. It is member of Class Basidiomycetes. *Pleurotus* contains almost 50 species (Hasan*et al.,* 2015; Jan et al., 2025).

Pleurotus djmor is chosen for the present examination, which is ordinarily known as oyster mushroom. It is being grown worldwide and is palatable. It is rich in protein (35–45% on dry weight basis). Medicinally, oyster mushroom is reported to diminish cholesterol levels in trial creatures (Hossain*et al.* 2003; Jan et al., 2025). It is a cellulose adoring organism and develops normally in the temperate and tropical woodlands on dead and rotting wooden logs or at times on drying trunks of deciduous or coniferous woods. It might likewise develop on rotting natural issue. The natural product bodies are little, develop independently or once in a while in groups, shellfish formed, once in a while grayish dark colored, stalk white and gills getting down to stalk (Chang *et al.*, 2004). Sreekala *et al.* (1997) reported that mesocarp fibers are left as a waste material after oil extraction, making extraordinary ecological issues. Therefore, monetary use of these fibers will be advantageous to the business cultivators and the nation altogether. Essentially, usage of agrarian waste as a media for the generation of mushroom is being assumed as a key job in diminishing the burn through and simultaneously making it valuable as a manure.

Material and Method

Seed source and research place

This study was carried out in the Department of Plant Pathology, University of Agriculture, Faisalabad during 2020. The pure culture of the mushroom was obtained from the mushroom lab, Institute of Horticultural sciences and was grown as the cultured medium followed by sub-cultured medium grown on Potato Dextrose Ager (PDA). Before culturing, the fruit bodies were immersed in the tap water to remove any type of debris on it.

Experimental plan

The culture obtained from the Department of Plant Pathology. The fruit bodies were cleaned with distilled water and dried in the shade on the newspaper. On drying, this media was again sterilized by using 1% sodium hypochlorite solution to remove the microorganisms. These fruit bodies were then chopped into small pieces by using the sterilized knife and with the help of scalpels, they were transferred to the growing media. The growing media containing the PDA was prepared in the sterilized petri dishes and fruit bodies were placed and incubated at the room temperature to grow the new fruit body of fungus. For the preparation of the spawns, sorghum grains were used as the growing media and these sorghum grains were sterilized by using the 1% sodium hypochlorite solution. Then these grains were sterilized by using the temperature treatment (how much temperature and also cite the paper of this protocol) and were half boiled. These grains were then placed in the plastic transparent bottles that were sterilized at 15 psi for 1 hour. The spawns were placed as 15, 25-, 35-, 45- and 55-grams rate and again autoclaved for the 1 hour at 30°C. These bottles were placed on the iron racks to allow the maximum mycelial growth. Once the bottles were filled with the white mycelia the bottles were ready for spawning in the different growth media i.e. cotton waste and rice cellulose waste.

Growth parameters

Number of flushes and number of bunches were counted at the time of harvest; however, the number of fruiting bodies per bunch were calculated by using the formula;

$$\text{Number of fruiting bodies per bunch} = \frac{\text{Total number of fruiting bodies}}{\text{Total number of bunches}}$$

Fresh and dry yield

The fresh yield of the mushroom was noted at the time of harvest by using the digital weight balance. For the measurement of the dry weight, the fruiting bodies per replication were noted by drying the mushroom bodies. For this purpose, the fresh produce was placed in the envelope and placed for 48 hours at 70°C. After that, the dry weight was measured and dry weight percentage was calculated by using the following formula and final reading was shown in percentage. (reference for this formula?)

$$\text{Yield (\%)} = \frac{\text{Total yield of all flushes from each replication (g)}}{\text{Substrate dry weight per replication (500 g)}} \times 100$$

Protein and total soluble solid contents

Total protein content was calculated as outlined by Bradford (1976). For this, 2g of the mushroom was chopped in already chilled mortal and placed in 7.6 pH phosphate buffer and centrifuged. The supernatant was then mixed with the Coosmassic blue solution (concentration of solution ?) and was rigorously stirred. The final reading was taken at 600 nm by using the spectrophotometer (mention model and company name) and a linear correlation was calculated and final reading was noted a Umg^{-1} per 100g fresh weight. On the other hand, the TSS was calculated by using the digital refractometer (mention model and company name) by putting a drop on its prism and direct reading was noted in the form of degree brix.

Biological efficiency

The biological efficiency of this was calculated by using the following formula in terms of percentage.

$$\text{Biological efficiency (\%)} = \frac{\text{Fresh Weight of Mushroom harvest}}{\text{Substrate Dry matter}} \times 100$$

Statistical Analysis

Data were analyzed using a two-way analysis of variance (ANOVA) in a completely randomized design (CRD) with factors being substrate type (cotton waste, rice cellulose waste) and spawn rate (15, 25, 35, 45, 55 g/bag). Means were compared using Tukey's HSD or Fisher's LSD test at $p < 0.05$. All analyses were performed using SPSS version 27. Data are presented as means ± standard error (SE) of four replications.

2. Result

Mycelial growth (days)First explain the results, don’t just make the table

Table 3.1 Different phases of completion of mycelia growth (days) grown on cotton waste and rice cellulose

Growing media	Treatment	Phases of mycelial growth (in days)			
		25%	**50%**	**75%**	**100%**
Cotton waste	T1 (15 spawn/bag)	3.6b	9b	15.6b	25.3b
	T2 (25 spawn/bag)	4.1c	10c	16.2c	22.1c
	T3 (35 spawn/bag)	4.6a	11a	17.0a	21.6a
	T4 (45 spawn/bag)	6.1e	12e	16.1e	20.0f
	T5 (55 spawn/bag)	6.5d	13d	19.d	24.3e
	LSD value	0.62	0.51	0.62	0.64
Rice cellulose	T1 (15 spawn/bag)	3.1c	8.2b	14.9bc	22.1b
	T2 (25 spawn/bag)	3.4bc	9.4c	15.1c	20.3c
	T3 (35 spawn/bag)	4.2b	10.2a	16.3a	19.6a
	T4 (45 spawn/bag)	5.7e	11.8e	15.1c	19.3e
	T5 (55 spawn/bag)	6.1d	12.1d	17.6d	22.1f
	LSD value	0.41	0.39	0.78	0.91

Pinhead formation (days)

The pinhead formation was stimulated by the application of the different growing media, but, the days for the pinhead formation of the *Pleurotus djamor* were significantly different in different growing media (Fig. 1). Maximum days for pinhead formation were taken by the 15 spawns per bag grown in the rice cellulose media as compared to the cotton waste treatments. The minimum days were taken by the 55 (33-days) spawn rate in rice cellulose media and cotton waste. As far as the treatments are concerned, 55 (33-days) spawn rates per bag was the best treatment that emerged first in both growing media followed by the 45, 35, 25 and 15 spawns per bag with 43, 56, 63 and 69 days, respectively (Fig. 1). Among the growth media, there were significantly less days required for the formation of the pinhead in cotton waste media as compared to the rice cellulose waste media**.**

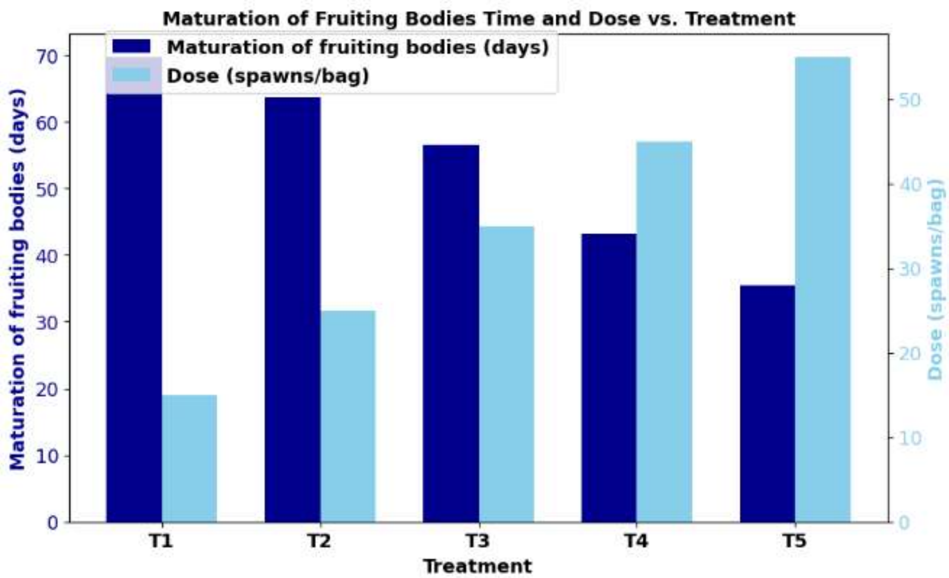

Fig. 1: Effect of spawn rates (15-55 g/bag) on days to pinhead formation of Pleurotus djamor on cotton waste and rice cellulose waste substrates. Bars represent means of four replications; error bars indicate SE.

Maturation of fruiting bodies (days)

The days for the maturation of fruiting bodies of the *Pleurotus djamor* from pinhead formation were significantly different in different growing media. The maturation of fruiting bodies was stimulated by using different growing media (Fig. 2). The maximum days taken for maturation of fruiting bodies were taken by the 15 spawns per bag treatment when grown in the rice cellulose media as compared to the cotton waste media. The minimum days were taken by the =55-spawn rate (5.01-days) both in rice cellulose media and cotton waste. As far as the treatments are concerned, 55 spawn rates per bag (5.01-days) was the best treatment that caused maturation of fruiting bodies earlier in both growing media followed by the 45, 35, 25 and 15 spawns per bag with 5.52, 5.60, 5.92 and 6.42 days, respectively (Fig. 2). Among the growth media, there were minimum days required for the maturation of the fruiting bodies in cotton waste media as compared to the rice cellulose waste media (Fig. 2).

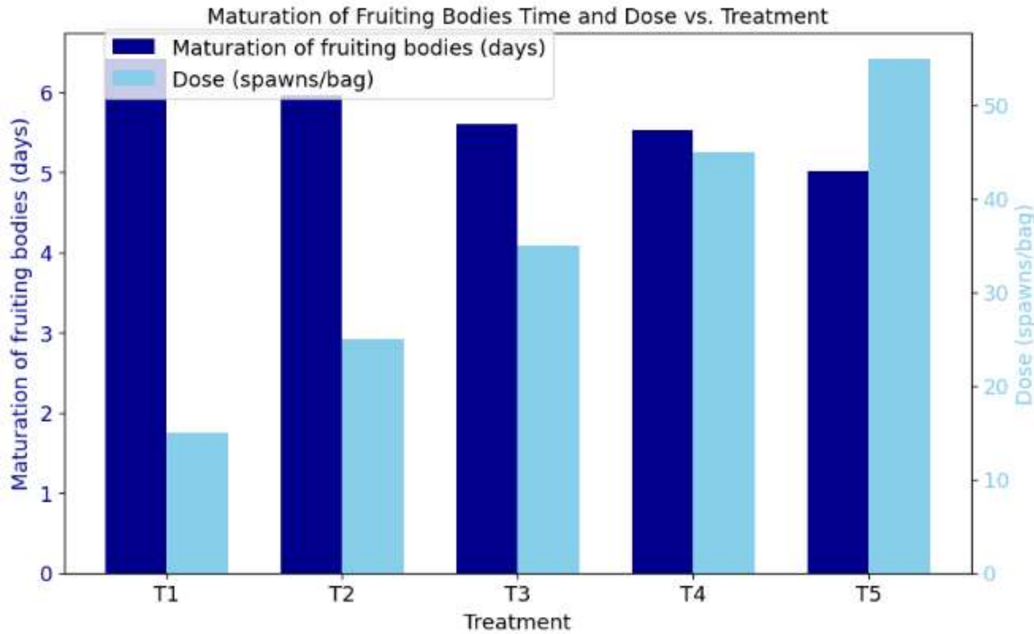

Fig. 2: Effect of spawn rates on days to maturation of fruiting bodies from pinhead formation

Fresh yield (%)

The fresh yield of the *Pleurotus djamor* from emergence to the maturation was significantly different in different growing media that was the mean value of four replications. The fresh yield was increased by the application of the different growing media (Fig. 3). There was maximum fresh yield in those grown in the 55 spawns per bag grown in the rice cellulose media as well as in cotton waste treatments; although, the cotton waste media was more effective as compared to the cellulose rice waste. The minimum fresh yield was obtained by 15 spawn rate both in rice cellulose media and cotton waste. As far as the treatments are concerned, 55 spawn rates per bag (41.62%) was the best treatment that had maximum fresh yield in both growing media followed by the 45, 35, 25 and 15 spawns per bag with 28.65, 24.56, 13.17 and 8.55%, respectively (Fig. 3). Among the growth media, there was significantly higher rate of the fresh yield in cotton waste media as compared to the rice cellulose waste media**.**

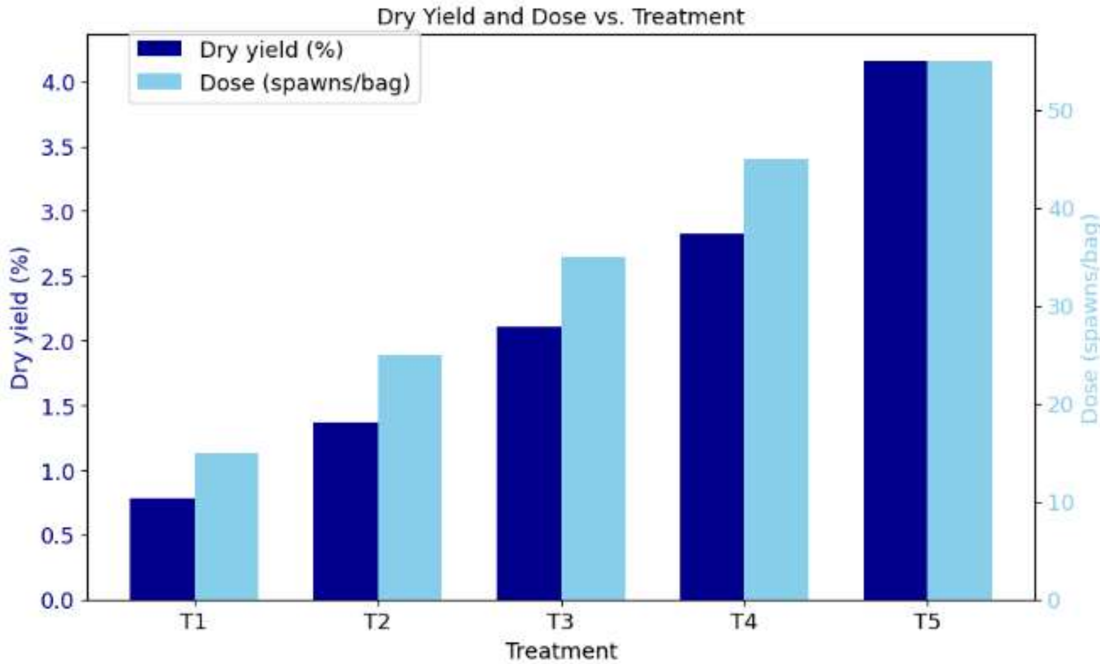

Fig. 3: Effect of spawn rates on fresh yield (%) of Pleurotus djamor.

Biological efficiency (%)

The biological efficiency of the *Pleurotus djamor* after harvest was significantly different in different growing media that was the mean value of four replications. The biological efficiency was increased by the application of the different growing media (Fig. 4). There was maximum biological efficiency in those grown in the 25 spawns per bag grown in the rice cellulose media as well as in cotton waste treatments; although, the cotton waste media was more effective as compared to the cellulose rice waste. The minimum biological efficiency was obtained by 55 spawn rate both in rice cellulose media and cotton waste. As far as the treatments are concerned, 25 spawn rates per bag (56.63%) was the best treatment that had maximum biological efficiency in both growing media followed by the 15, 45, 35 and 55 spawns per bag with 51.78, 42.75, 37.02 and 34.02% biological efficiency, respectively (Fig. 4). Among the growth media, there was significantly higher rate of the biological efficiency in cotton waste media as compared to the rice cellulose waste media

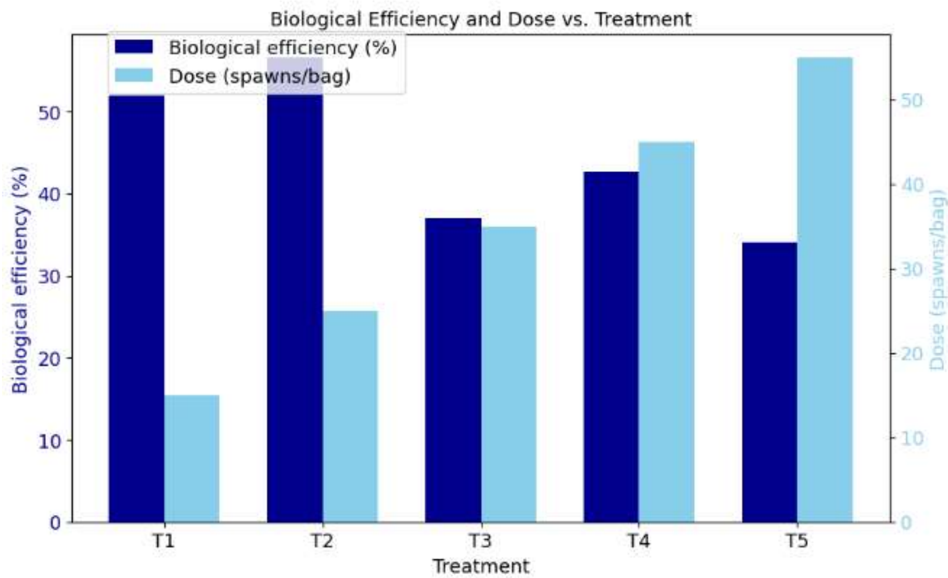

Fig. 4: Effect of spawn rates on biological efficiency (%) of Pleurotus djamor.

Conclusion

The present study demonstrates that both substrate type and spawn rate substantially affect the growth and yield performance of Pleurotus djamor. Among the tested treatments, cotton waste proved to be a more effective substrate than rice cellulose, supporting faster mycelial colonization, early fruiting, and higher fresh yield. The highest yield and quality attributes were achieved at a 55 g spawn rate per 500 g substrate bag, while biological efficiency was greatest at a 25 g spawn rate. These results suggest that a higher spawn rate accelerates substrate colonization and fruiting, whereas moderate spawn rates enhance resource utilization efficiency. Consequently, the use of cotton waste supplemented with 55 g of spawn represents an optimal combination for maximizing Pleurotus djamor productivity. This approach not only improves mushroom yield but also promotes sustainable management of agricultural residues, offering a low-cost, eco-friendly avenue for rural entrepreneurship and waste valorization in mushroom cultivation systems.

Acknowledgment

The author expresses sincere gratitude to **Professor Dr. Nasir Ahmed Khan**, Department of Plant Pathology, University of Agriculture, Faisalabad, for his invaluable guidance, supervision, and continuous support throughout the course of this research. His expert advice, encouragement, and constructive suggestions were instrumental in shaping this work to completion.

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