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Evaluation of Various Substrates on Morphological Character and Yield of *Pleurotus djamor*

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A B S T R A C T

Mushroom is cultivated across the world which is highly nutritious and pharmaceutically useful for the human. Oyster mushroom is growing on agriculture waste material by using argo-cellulosic for growing to easily decompose agricultural waste material. Substrate is *Triticum aestivum* as control and other substrate is *Oryza sativa, Lens culinaris* and *Cicer areitum* is taken to grow *Pleutotus djamor*. morphological evaluation of growing mushroom on substrate result is *Oryza sativa*, *Lens culinaris* and *Cicer aeritium* as mycelium initiation, spawn run days and pinhead appearance and siphonophore's formation and harvesting and biological efficiency as well as fresh and dry weight of the taken mushroom gives *Oryza sativa*, *Lens culinaris* and *Cicer aeritum* respectively. Mean values under the same category that bear different superscript letters are significantly different ($\alpha < 0.05$).

***Key words*:** *Pleurotus djamor*, Sporophores, Cultivation, Biological efficiency

A mushroom or toad stool is a fungus's fleshy, spore-bearing fruiting body that grows above ground, on soil, or on its feeding supply. Mushroom are rich in nutrients and B Vitamin containing niacin, riboflavin and pantothenic acid and play important role in nervous system. Mushroom are low in calories, sodium and free from fat, cholesterol [1]. Mushroom is taken for their texture, flavor and nutritional values and this is potent of 30% proteins, 56% carbohydrates, vitamins (potent in pyridoxine, folic acid, cobalamin, thiamin e, niacin, riboflavin, nicotinic acid, pantothenic acid, ascorbic acid and vitamin D&B and other vitamins, such as ergosterol, biotin and tocopherols), minerals (iron, potassium, phosphorus, calcium, copper), amino acid (rich in lysine and leucine) and low-fat content (2-8%). Mushroom is preferred as food for treat against hypertension, arthrosclerosis, diabetes and obesity etc. Mushroom interestingly have vital role in health benefits and its development of new biological remediation techniques and it also generate revenue income [2]. Mushrooms are increasingly being recognized as important food products for their significant role in human health, nutrition and disease. Several species of mushrooms are of great importance because of their medicinal properties, for example, they are active against hypercholesterolemic conditions, hypertension, diabetes, cancer and other infections [3]. Cultivation of the oyster mushroom, *Pleurotus* spp., has increased greatly throughout the world and commonly grown on pasteurized agro wastes. It can be cultivated on a wide variety of lignocellulosic substrates, enabling it to play an important role in managing organic wastes whose disposal is problematic. Mushroom cultivation is a simple, low cost and environmentally friendly technology for the utilization of rural and agro-industrial residues. The substrate used for the cultivation of one such species is pink oyster mushroom, *Pleurotus djamor* var. roseus, which is becoming important as this is an unfamiliar edible mushroom and can be cultivated easily throughout the year [4]. Substrates affects production, yield, and nutrients content in oyster mushrooms. Pink oyster mushroom is grown on various substrate as g wheat, paddy or oat straw, sawdust, sugarcane bagasse, date palm fiber, rice and wheat bran and especially on sawdust (Jackfruit, mango, olive, rain tree, eucalyptus and mixed of all) supplemented with 30% wheat bran during growth. ink oyster mushroom, which is lignocellulose decomposer and it has a light to dark pink cap and intensity depends upon the strain and growing conditions. Primordia and young mushrooms are bright pink at the initial stage and turned to pale pink at its mature stage of growth its classification as family Pleurotaceae, the order Agaricales, and the class Agaricomycetes and the division Basidiomycetes [5]. Research is commenced on medicinal macro fungi in 1960 in Japan, China and Korea [6]. *Pleurotus djamor* commonly called Oyster mushroom is commercial cultivated over the world and India, due to the unique ability to produce extra cellular lignocellulolytic enzymes and allowing then to grow in a wide range of agriculture waste [7]. Yield outcoming from 1 kg per bags is very less and its solution is that pink oyster mushroom is grown on different agriculture waste substrate for increasing production of cultivated mushroom. I have grown pink oyster mushroom on different cellulosic substrates for higher

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production from various substrates as paddy, lentil and gram counter with wheat substrate as control. Mycelia of mushroom have extracellular enzyme can utilize and degrade the lignocellulose wastes and they play important role to reduce pollution and also restoration of damage environment [8].

MATERIALS AND METHODS

Study area and experimental material-The research was carried out in the Laboratory of Mushroom Biology, Department of Botany, Dr. H.S.G. (A Central), University of Sagar (MP), India, from 15 June, 2021. To 15, November, 2021. Varied organic substrates, namely *Triticum aestivum*, *Lens culinaris*, *Cicer arietinum* and *Oryza sativa* were evaluated for cultivation of pink oyster mushroom. The substrates were collected from the research field at the aforementioned department. Pure culture of pink oyster mushroom was procured from the ICAR, Solan Research institute, Himanchal Pradesh, India. The culture code of *Pleurotus djamor* var. *roseus*. Pure culture preparation and production condition- For 7 days, mushroom cultures were cultivated on Potato Dextrose Agar (PDA) media using standard method [9]. Slants were prepared aseptically and the fungus was sub cultured by transferring a piece of 4 mm by 4 mm fleshy tissue to fresh PDA slant. The inoculated slants were incubated at 25 °C for 7 days. Once the slants were completely covered with fungal mycelia, the culture was used for spawn preparation.

Spawn preparation

Mushroom cultivation technology, the inoculums is also known as the spawn [10]. For spawn preparation, 4 kg of partially cooked clean sorghum grains were used. Excess of the water was removed and grains were cooled to room temperature. The cooled grains were spread on sterile table to bring the grain moisture to approximately 50%. It was then mixed with 2 percent calcium sulphate (mention the company name). The mixture was then filled in a one litre bottles to autoclave at 22 psi and 121 °C for 1hour. Sterilized bottles were removed from the autoclave while still hot and shook to minimize grain clumping. Sterilized bottles were inoculated with 5 mm by 5 mm agar piece of subculture slant of *Pleurotus djamor* and briskly shaken. The spawn was incubated in dark at 24 ± 1 °C for 20 days**.**

Substrate preparation and spawning

Triticum aestivum straw, *Lens culinaris* branch shreds, *Cicer arietinum* whole plant shreds and *Oryza sativa* straw were sundried for about 20 days and measured for difference in weight, until no moisture could further loose. 10 kg of dried substrates were transferred to individual tanks filled with 5% carbendazim and 35 % formalin for 18 hours. Tank was covered with polythene sheet to prevent the evaporation. The straw was taken from the tank and left for 2-3 hours to drain the excess water, until 80 % moisture remained. The substrates were filled in polythene bags (how many kgs) and aseptically inoculated with spawns of *Pleurotus djamor* as intermittent 5 layers. The bags were then punched with 8-10 holes for aeration [11].

Cultivation condition and cropping system

The bags were kept under dark condition in crop rooms. Spawn run in crop room had temperature (22-26 °C) and relative humidity (80-90%) during spawn run. Humidity was maintained by spraying water three times a day. After the mycelial growth in bags were abundant, the substrate turned into compact mass with whitish mycelial growth or pinheads visible. The windows were kept open for 1-2 hours during sporophores development to control fresh air and eliminate CO_2, keeping the relative humidity within the crop room at 80-90 percent.

Data collection

The mushroom's growth and development were tracked on a daily basis. The time (in days) necessary from inoculation to completion of mycelium running, the time required from opening the plastic bags to pinhead formation, and the time required from opening the plastic bags to first round harvesting were all documented. Before each harvest, growth metrics such as stipe length (cm), stipe diameter (cm), pileus diameter (cm), and pileus thickness (cm) were measured using a slide calliper. At harvest, yield parameters such as the number of fruiting bodies per bunch and total fresh weight (g) of mushroom were also recorded. Mature fruiting bodies (white in colour, with up curving pileus) were gathered by slicing the base with a sharp blade slightly above the surface of the substrate. During the experiment, two rounds of mushroom harvesting were conducted across all substrate types. Yield and biological efficiency were calculated to assess the growing performance of mushrooms on various substrates. As a result, biological yield (g) was calculated by weighing the whole cluster of fruiting bodies without removing the stalk bases, while economic yield (g) was calculated by weighing all of the fruiting bodies on a substrate after removing the stalk bases. Finally, biological efficiency (percentage) was determined as follows:

$$\%BE = \left(\frac{FWm}{DWs}\right)*100$$

Where;
BE denotes Biological Efficiency (%),
FWm denotes total fresh weight (g) of mushroom yield across all flushes, and DWs denotes substrate dry weight (g).

Statistical analysis

Then, using SPSS version 20, an analysis of variance (ANOVA) was performed, and the mean values of all parameters, as well as the standard errors of each parameter, were separated using LSD at a 5% level of significance.

RESULTS AND DISCUSSION

Time elapsed for mycelial running, pin-head formation and maturity of fruiting body

Time elapsed for mycelial initiation, mycelial running, pin-head formation and maturity of siphonophores outcomes on the overall time required for mycelial initiation, running, pin-head formation and maturity of fruiting bodies are illustrated in (Table 1). Number of days to mycelium initiation was almost similar in all substrates. Mycelial growth was faster on *Oryza sativa* and *Lens culinaris* substrate than on wheat straw (12 days) and *Cicer arietinum* (12.33 days). The period of pin-head formation varied with substrates, ranging from 14 to 21 days after spawn seeding. Pin-head formation occurred comparatively faster in *Oryza sativa* substrate (14 days), followed by *Lens culinaris* (15 days), *Cicer arietinum* (16 days); while it took relatively longer time in *Triticum aestivum* (21 days). *Lens culinaris* substrate and *Oryza sativa* substrate had higher mycelia density as compared to that of wheat straw and *Cicer arietinum* substrate. The time required for maturing of sporophore ranged from 15.33 days for *Oryza sativa* and 18 days and (for *Lens culinaris* and *Cicer arietinum*) to 24.33 days (for wheat straw).

Table 1 Time elapsed for mycelia running, pin-head formation and maturity of fruiting bodies of mushroom under different substrates. Values are given in average of three replicates

Characteristics	*Triticum aestivum*	*Oryza sativa*	*Cicer arietinum*	*Lens culinaris*
Mycelium initiation no. of days	2.33 ± 0.3[a]	2 ± 00 [a]	2 ± 00 [a]	2 ± 00 [a]
Complete mycelium of spawn run no. of days	12.33 ± 0.33 [a]	11.67 ± 0.33 [b]	12.67 ± 0.33 [c]	12.67 ± 0.33 [d]
Pinhead Appearance no. of days	21 ± 0.58 [a]	14 ± 0.00 [b]	16.67 ± 0.33 [c]	15.67 ± 0.33 [d]
Sporophores appearance no. of days	24.33 ± 0.67 [a]	15.33 ± 0.33 [b]	18 ± 0.58 [c]	18.33 ± 0.33 [d]

Mean values under the same category that bear different superscript letters are significantly different (α < 0.05). Superscript similar alphabet has same means

Biological and economic yield

Results of the yield components (yield attributes) of oyster mushroom grown in each substrate are presented in (Table 2). Accordingly, it was found that the product from *Oryza sativa* substrate had a relatively better growth in terms of perimeter, height and diameter of the pileus, and stipe length. It was observed that the number of fruiting bodies was significantly higher in the culture of paddy substrate from other substrates. The lowest number of fruiting bodies was recorded in *Cicer arietinum* substrate. Number of days required for first, second and third harvest, fresh weight and dry weight also differed from each other. Matured fruiting bodies of oyster mushroom were harvested and weighed to determine biological and economic yield. The largest yield was harvested from *Oryza sativa* substrate, followed by *Lens culinaris*; while, the least was obtained from *C. arietinum*. Further separation of the mean yields was also made using the LSD test to see whether there is significant yield difference among the substrates. first, second and third harvesting no of days, fresh weight and dry weight is also significantly differ from each other.

Table 2 Biological and economic yield first, second and third harvest and total harvest graph biological yield. Values are given in average of three replicate

Characteristics	*Triticum aestivum*	*Oryza sativa*	*Cicer arietinum*	*Lens culinaris*
Mycelium initiation no. of days	26.67 ± 0.3 [a]	17 ± 0.00 [b]	20.67 ± 0.33 [c]	20 ± 0.00 [d]
Complete mycelium of spawn run no. of days	31 ± 0.58 [a]	20.33 ± 0.33 [b]	28.67 ± 0.33 [c]	23 ± 0.33 [d]
Pinhead Appearance no. of days	38.67 ± 0.33 [a]	23.67 ± 0.33 [b]	34.33 ± 0.33 [c]	28.33 ± 0.33 [d]
Sporophores appearance no. of days	140.33 ± 4.5 [a]	238 ± 7.57 [b]	69.67 ± 2.60 [c]	213 ± 3.51 [d]

Mean values under the same category that bear different superscript letters are significantly different (α < 0.05). Superscript similar alphabet has same means

Biological efficiency

As shown in (Fig 1), results of the biological efficiency varied significantly among the substrates used. The highest percentage of biological efficiency was obtained from *Oryza sativa* substrate (23.80gm); while the least was observed in *Cicer arietinum* substrate (21.30gm).

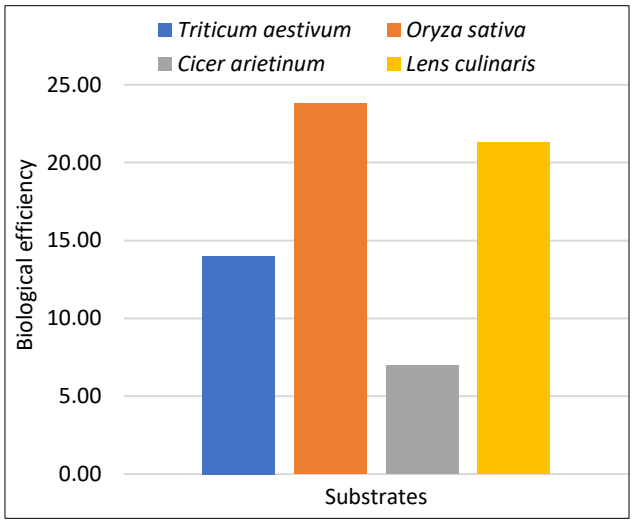

Fig 1 Comparison of biological efficiency of mushroom yield using different substrate

Triticum aestivum and paddy are the staple crops, producing ample of stover yield in kharif and rabi seasons. Mushroom cultivation over such agriculturally based waste products add up in substance or subsidiary agriculture. Since most of the agricultural land is rain water irrigated, not all every farmer can choose wheat or paddy in their fields. Wheat and paddy are shown to have better mushroom yield; hence wheat straw was chosen as control. *Lens culinaris* and *Cicer arietinum* are crops of choice in regions of lesser irrigation. Agricultural waste material resulting from these crops are generally used as fuel for cooking or burnt. This cellulosic biomass has the potential to be used by people to grows mushroom. The mushroom cultivation in rural regions rely on agriculture cellulosic waste material produced by the farmers themselves after the harvest of season. Commercial production of mushroom would require a seamless supply of such agricultural waste, but marginal farmers don't have the infrastructure to stock up the raw material. Hence, the alternative substrate in agriculture with potential to grow mushroom is a requisite to improve rural economy in developing countries.

Different agricultural wastes were tested in the experiment to test the growth performance of different substrates. *Oryza sativa* mycelium ran normal and good amount was filled with bag with enormous fruiting. *Lens culinaris* mycelium grew slow and filled less in comparison to control. Bag filled with mycelium completely gives very good amount maturing pink fruiting body. This is probably due to high carbon to nitrogen ratio in hard woody substrates [12]. The pink color disappeared as the maturing commenced. *Cicer arietinum* substrate is filled with mycelium is slow as control and fruiting body after maturing color turn into pink and flush was obtained in small quantity. First second and third harvest was also completed to take more time. Fresh weight is also less produced and biologically economic yield is also poor obtained comparison to control and other substrate. *Oryza sativa* substrate took lesser number of days to generate mature fruiting body with highest biological efficiency. In comparison, *Lens culinaris* and *Cicer arietinum* did not exhibit a desirable biological efficiency. In this research it was found that good biological efficiency was in the order *Oryza sativa* > *Lens culinaris* > *Cicer arietinum*. Many substrates from agricultural sources have previously been proven to be useful for production

of pink oyster mushroom with good biological efficiency [13]. There was a missing data of use of *Cicer arietinum* as a carbon source in pink oyster production. Though it was found to be not desirable for pink oyster production due to lower yield and biological efficiency.

CONCLUSION

Mycelium initiation, complete mycelium of spawn run, pinhead appearance and sporophores appearance and first, second and number of days for third harvesting is increasing order is *Oryza sativa* and comparatively same both *Lens culinaris* and *Cicer arietinum* and fresh and dry weight and biological efficiency is highest in *Oryza sativa*, *Lens culinaris* than *Cicer arietinum* respectively, therefore *Oryza sativa*, *Lens culinaris* and *Cicer arietinum* agriculture waste substrate is suitable for the cultivation of pink oyster mushroom. It is gives low economically yield but it is also used as fodder and protein feed for animals. Woody agriculture substrate decompose slowly, but by growth of mushroom aid in breakdown and offer more sustainable facet for the research and further analysis of agriculture substrate.

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