

# Characterizing the Growth Kinetics of Oyster Mushroom Through Substrate Supplementation and Environmental Control

Khurram Shahzad, Hamza Aslam, Shahid Hussain\*

Department of Biotechnology, Kohsar University, Murree, Pakistan.

## ABSTRACT

**Background:** Oyster mushrooms (*Pleurotus ostreatus*) are valued for their nutritional, economic, and ecological benefits.

**Objective:** This study evaluated the effects of carbon and nitrogen supplementation on oyster mushroom growth using wheat straw as the base substrate.

**Methodology:** Four treatments were compared: (1) wheat straw alone (control), (2) wheat straw plus molasses, (3) wheat straw plus sugar solution, and (4) wheat straw exposed to sunlight without daily watering. Both molasses and sugar supplementation accelerated mycelial growth (15 vs. 18 days), pinhead formation (18–19 vs. 22–23 days), and fruiting body maturation (21–22 vs. 25–26 days) compared to the control.

**Result:** Sunlight-exposed substrates exhibited delayed and smaller flushes. Fourier Transform Infrared (FTIR) analysis of dried mushrooms confirmed the presence of hydroxyl, carbonyl, amine, and polysaccharide functional groups. Optimizing substrate supplementation can enhance the productivity and profitability of oyster mushroom farming, particularly for small-scale or resource-limited growers.

**Conclusion:** Future work should explore additional nutrient additives, environmental variables, and the mushrooms' bioactive compounds to further improve cultivation efficiency and nutritional value.

### Keywords

Growth Kinetics, Oyster Mushroom, Characterization.

### \*Address of Correspondence

shahid.hussain@kum.edu.pk

### Article info.

Received: March 12, 2025  
Accepted: June 04, 2025

**Cite this article:** Shahzad K, Aslam H, Hussain S. Characterizing the Growth Kinetics of Oyster Mushroom Through Substrate Supplementation and Environmental Control. *RADS J Biol Res Appl Sci.* 2025; 16(1):27-32.

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## INTRODUCTION

Mushrooms are umbrella-shaped fruiting plants, consisting of a cap (pileus) and a stalk (stipe). They can be edible or poisonous and have rich protein, vitamins, mineral content, and unique flavor. Almost 3,000 edible species, over 700 are recognized for medicinal properties<sup>1</sup>. Mushroom cultivation, the second most significant microbial technology after yeast, is gaining popularity globally for its economic, nutritional, and therapeutic benefits. Typically, mushrooms are composed of 90% water and 10% dry

matter, making them a nutrient-dense food source<sup>2-4</sup>. With the cultivation of various mushrooms, mushroom culture has diversified significantly. Edible mushrooms can colonize and break down many lignocellulosic substrates and agricultural, forestry, and food-processing wastes, further enhancing their ecological and economic value<sup>4</sup>.

Some mushrooms are easily cultivable and have significant global markets. Over 200 species are collected from the wild and used in traditional medicine, particularly in the Far East. Among cultivated mushrooms, *Agaricus*

*bisporus* is the most widely farmed species worldwide<sup>4-6</sup>. Mushroom cultivation requires careful monitoring and regulation of growing conditions. Temperature and relative humidity are critical factors that significantly impact mushroom production during all stages of growth, including spawning, fruit induction, and harvesting<sup>7</sup>. Mushroom cultivation can reduce poverty and improve livelihoods by providing a high-yield, nutritious food source and a stable income. It is an accessible activity for rural and peri-urban farmers, as it requires minimal land, low capital investment, and uses clean agricultural residues as substrates. Mushrooms can be grown in simple, temporary shelters with little maintenance, making them ideal for small-scale or part-time farming<sup>8</sup>.

The oyster mushroom (*Pleurotus ostreatus*), a member of the family *Agaricaceae* and phylum *Basidiomycota*, originated in China and is now cultivated worldwide, except in the Arctic regions of the northwest Pacific due to the extreme temperature. During World War I, Germany developed large-scale cultivation techniques for *P. ostreatus* as part of efforts to address food shortages. Today, the species is widespread, including across Poland<sup>9</sup>. Whereas, in the Czech Republic and Slovakia, *Pleurotus ostreatus* is commonly used as a meat alternative, while in many other countries, particularly in Asia, it is considered a delicacy. The *Pleurotus* genus has been extensively studied and cultivated worldwide for various purposes, thriving on a wide range of lignocellulosic substrates<sup>10</sup>.

*P. ostreatus* requires minimal environmental control for cultivation, as their fruiting bodies are typically resistant to diseases and pests. Additionally, their cultivation process is straightforward, low-cost, and efficient. Nutritionally, oyster mushrooms are a rich source of vitamin C, chlorine, selenium, iron, and calcium. Furthermore, they are recognized for their therapeutic potential, particularly in the treatment of cancer and diabetes, due to their bioactive compounds<sup>11,12</sup>. The success of mushroom cultivation is influenced by several factors, including temperature, humidity, and the sterility of the substrates, which may interact in complex ways. However, excessive cultivation can lead to significant losses in yield, primarily due to increased susceptibility to pests and diseases. Proper management of these factors is essential to optimize

productivity and minimize crop loss<sup>13,14</sup>. The fruiting bodies of oyster mushrooms are rich in mineral elements, with potassium (K), phosphorus (P), sodium (Na), calcium (Ca), and magnesium (Mg) being the major constituents. Trace elements such as copper (Cu), zinc (Zn), iron (Fe), molybdenum (Mo), and cadmium (Cd) are also present in smaller quantities. Among these, *P. ostreatus* has particularly high concentrations of copper (Cu), iron (Fe), potassium (K), magnesium (Mg), phosphorus (P), zinc (Zn), and sodium (Na)<sup>15</sup>. However, while mushrooms contain carotenoids, which have the potential to act as precursors to retinol (vitamin A), their concentration of these compounds is comparatively low when compared to plant sources. As a result, mushrooms are not a significant source of carotenoids<sup>16</sup>. Mushrooms seem to be relatively rich in riboflavin, niacin and ergocalciferol. Available data on ergosterol, the provitamin of ergocalciferol. The relatively high ergosterol content could be of significance for individuals with a limited intake of ergocalciferol from foods of animal origin, e.g. for vegetarians and vegans. The occurrence of carotenoids, including those which can act as precursors of retinol, is limited in mushrooms compared to plants.<sup>17-20</sup>.

This study aimed at culturing of oyster mushroom, including four sets of major experiments consisting supplementation of the gradual variation in the carbon and nitrogen sources under selected environmental conditions.

## MATERIALS AND METHODS

### Materials

Molasses, wheat straws, sugar, gelatin, and water were utilized as supplemental sources.

### Preparation of Substrates

The collected wheat straws were converted into smaller pieces, soaked in water, and boiled for about 5 h. The substrate absorbed 70 to 80% of the water, and excess water was removed. The spawn of mushrooms formed in sterile conditions. Then a spore of mushroom was taken in a separate plate and autoclaved with gelatin and other carbon sources. After a few days from the spore, mycelial growth appeared. The grown mycelium was added with boiled grains of wheat for the preparation of seeding for oyster mushrooms. The substrate was mixed with mushroom spawn and put in the polythene bag. Firstly, substrate was added in the

polythene bag, then spawn was added, and then more substrate, and vice versa. After mixing the substrate and spawn, the polythene bag was closed. Small cuts were created for aeration and placed in a dark and humid place. The water was showered daily on the sample.

In the second experiment, in addition to the above procedure, a molasses solution was provided to the substrate to get a high yield of mushrooms. For the third experiment, a sugar solution was provided for mushroom culturing. Moreover, for the fourth experiment, the substrate and spawn were directly exposed to sunlight without watering daily.

### Characterization Studies

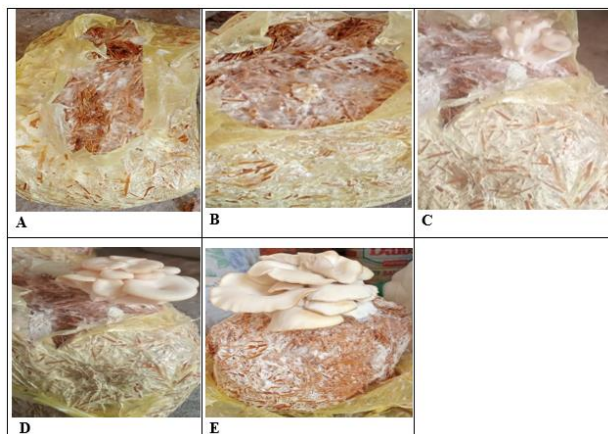
Oyster mushrooms were dried in the oven for 4-6 h and then converted into fine powder. This dried powder was analyzed by Fourier Transform Infrared (FTIR) Spectroscopy.

## RESULT

After several days, mycelium grew fully and made a white appearance on the substrate. After 3 to 4 days of mycelium growth, a small branch of mushroom started growing out of the substrate from the cut that was made on the polythene bags. Soon after 3 or 4 days, these branches became a full mushroom, and these branches are called flushes. In all experiments, flushes are equal in size except for the fourth experiment. In experiment four, the branches start growing after 25 days, and these branches are smaller than those of other experiments.

### Growth of Mycelium on Wheat Straw

After adding the spawn in the wheat straw, the mycelium grew all around the wheat straw after 10 days. The white, web-like structure appeared on the whole substrate. Due to close packing by the mycelium, it became hard. Small branches appeared on the substrate after 4 to 5 days. After 8 days, the oyster mushroom fruiting body reached full maturity in size. At this stage, the fruiting body of the oyster mushroom was fully developed, displaying its characteristic cap, gills, and stem structure. The growth timeline for the oyster mushroom, from colonization of the substrate to full fruiting body development, was typically rapid, and the mushrooms reached their mature size in a relatively short period under optimal conditions (Figure. 1 and Table 1).



**Figure 1.** (A) Mycelium growth in the substrate, (B) Growth of small branches on the first day, (C) Third-day growth of small branches of oyster mushroom, (D) Fifth-day growth of branches of oyster mushroom, (E) Full growth of oyster mushroom.

### Growth of Mushroom with Molasses

Molasses was used as an additive nutrient for stimulation of growth. Due to molasses, it had reduced the growth period of mushrooms. Molasses is a source of nitrogen and sugar that are causative agents for stimulation of growth. In normal growth, mycelium grew in up to 18 days, but in the case of molasses, it grew in only 15 days. In normal growth, a pinhead was formed in 4 to 5 days after full mycelium growth, but in case of molasses, it formed in 3 to 4 days. In normal growth, a full fruiting body was formed in 3 to 4 days after pinhead formation but in case of molasses, it grew in 1.5 to 2 days. The result of sugar solution and molasses was the same. Because both are source of sugar that were the causative agent for the stimulation of growth (as shown in Figure 2 and Table 1).

### FTIR Analysis

The FTIR spectrum showed more than five band regions that showed complexity (Figure 3). The bands were ranging from  $3425.58\text{ cm}^{-1}$  to  $640.37\text{ cm}^{-1}$ . The broad peak in the region around  $3400\text{-}3200\text{ cm}^{-1}$  indicated the hydroxyl groups (OH) and -NH group for protein. The sharp peak in the region around  $1630\text{-}1800\text{ cm}^{-1}$  showed the presence of the C=O functional group. This narrow peak showed the presence of aldehydes, ketones, esters, and carboxylic acid. A sharp peak below the  $3000\text{ cm}^{-1}$  showed the presence of alkane C-H. The peaks above  $3000\text{ cm}^{-1}$  showed the presence of a double bond C=C. A little peak

appeared in the region between 2000 and 2200  $\text{cm}^{-1}$ , showing the presence of a triple bond in the compound. The band region from 1450  $\text{cm}^{-1}$  to 1276  $\text{cm}^{-1}$  represents the -CH group for proteins and the -P=O phosphodiester group for nucleic acids and phospholipids. The band region from 1153  $\text{cm}^{-1}$  to 1092  $\text{cm}^{-1}$  represents the -C-O group

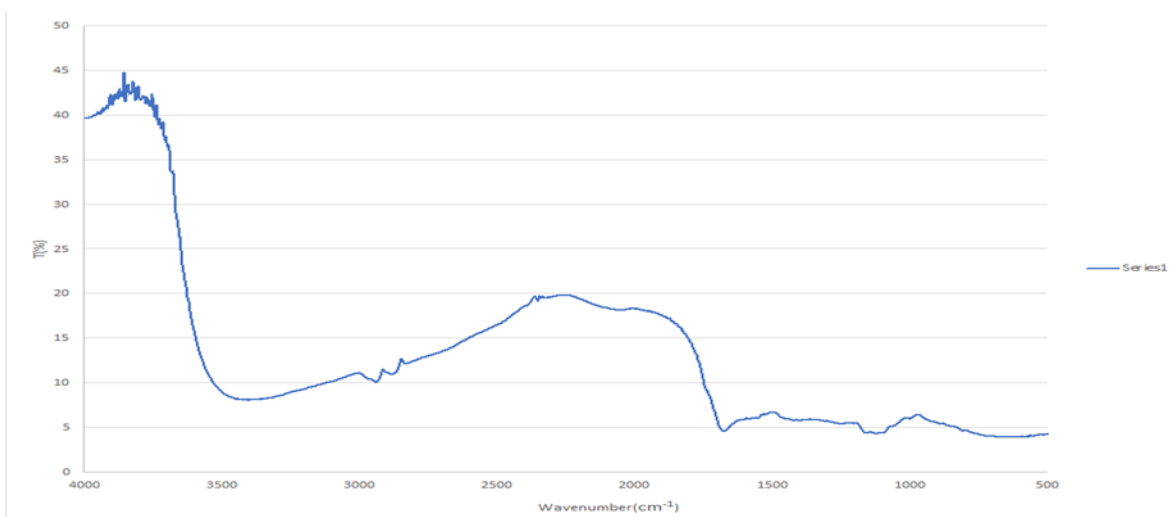
from the pyranose ring of  $\beta$  (1 $\rightarrow$ 4) glucans and the -C-O group from  $\beta$  (1 $\rightarrow$ 3) glucans. The band region from 995  $\text{cm}^{-1}$  represents the group corresponding to  $\alpha$ -glycoside. The band region from 787  $\text{cm}^{-1}$  to 581  $\text{cm}^{-1}$  represents the -C-O group from polysaccharides (Figure 3).



**Figure 2.** Full growth of oyster mushroom with molasses (left) and full growth of oyster mushroom with sugar solution (right).

**Table 1. Difference Between Normal Growth, with Molasses, and with Sugar Solution.**

Substrate	Spawn Running/ Mycelium Growth	Pinhead Formation	Fruiting Body Formation
Wheat Straw	18 days	22 to 23 days	25 to 26 days
Wheat Straw + Molasses Solution	15 days	18 to 19 days	21 to 22 days
Wheat Straw + Sugar Solution	15 days	18 to 19 days	21 to 22 days



**Figure 3.** The FTIR spectrum of oyster mushroom.

## DISCUSSION

In this study, oyster mushrooms were locally cultivated using wheat straw as a substrate in combination with molasses and sugar solution. Through an estimation, one kg of production cost is around Rs 80, and it could be sold in the market at the rate of Rs 380. Cultivation of mushrooms on a large scale can provide the population with an affordable protein source. These can also replace animal protein for people who cannot afford it or are allowed to eat due to medical reasons. Findings from this study demonstrate the significant impact of substrate composition and nutrient supplementation on the growth rate and yield of oyster mushrooms (*Pleurotus ostreatus*). The results indicated that adding molasses or sugar solution to the wheat straw substrate accelerates the growth of mycelium, pinhead formation, and fruiting body development, supporting previous studies that highlight the importance of nutrient sources in mushroom cultivation<sup>21,22</sup>. This suggests that the additional sugar sources in molasses and sugar solution serve as a readily available energy source for the mycelium, facilitating faster growth. The results align with studies that report enhanced mycelial growth and reduced cultivation periods with the addition of sugars and nitrogen-rich compounds<sup>23,24</sup>. However, the FTIR analysis provided valuable insights into the biochemical composition of the oyster mushrooms. The presence of multiple functional groups, such as hydroxyl, carbonyl, and amino groups, suggests that oyster mushrooms contain complex compounds, including proteins, lipids, and carbohydrates. The FTIR results support the potential medicinal and nutritional value of oyster mushrooms, confirming their role as both a functional food and a source of bioactive compounds<sup>25,26</sup>. Moreover, the results suggest that oyster mushroom cultivation can be optimized further by exploring other nutrient supplements or variations in environmental conditions, such as light exposure and humidity levels, to further enhance growth rates and fruiting body size.

## CONCLUSION

These findings highlight the potential for optimizing substrate composition and cultivation conditions to enhance mushroom yield, quality, and profitability. This will contribute to sustainable agricultural practices and food

security. Future research should explore the use of other nutrient additives, and environmental factors as well as their medicinal properties.

## CONFLICT OF INTEREST

None.

## ACKNOWLEDGEMENT

None.

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