ISSN 2348-6201

Growth Performance and Cultivation of Four Oyster Mushroom Species on Sawdust and Rice Bran Substrates

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ABSTRACT

Pleurotus species are progressively becoming important edible mushrooms worldwide and it is of importance in choosing suitable substrates in a given place to grow them. The study provides basic and valuable information on mycelial growth rates and yield efficacy of four different *Pleurotus* spp. The highest average mycelium growth of 1.29 mm/day followed by 1.17 mm/day was observed in the bags of *P. salmoneostramineus* and *P. ostreatus* grey, respectively. Similarly, the highest cumulative growth of 12.90 cm/day and 11.74 cm/day was recorded in the 30 day old bags of *P. salmoneostramineus* and *P. ostreatus* grey, respectively. There was a significant increase ($P < 0.05$) of about three fold in the pin head formation of *P. ostreatus* grey (9 days) compared to other mushroom species (2-3 days). *P. salmoneostramineus* showed a vigorous growth in the fruiting body and the harvesting time required was just 5.67 days. It is clear from the result that there is no correlation between the rate of mycelia growth and pin appearance. The yield varied significantly due to the effect of mycelial growth on the substrate. The highest first flush and number of fruiting bodies was achieved in the yellow oyster followed by *P. ostreatus* grey and *P. salmoneostramineus*. In conclusion, sawdust and rice bran could be a promising substrate for linear mycelial growth of *P. ostreatus* species.

Indexing terms/Keywords

Mycelium Linear Growth, Oyster mushrooms, sawdust, rice bran, bags.

Academic Discipline And Sub-Disciplines

Biotechnology

SUBJECT CLASSIFICATION

Cultivation

TYPE (METHOD/APPROACH)

Experimental.

INTRODUCTION

The ever growing need of producing cheap nutritious food, led to the fast development of the mushroom cultivation industry [1]. The current market is eagerly looking for the introduction of new mushroom products through the cultivation of exotic species, which provides an array of novel choices for the consumer and promotes social development [2, 3]. The large quantities of agro-industrial wastes that are produced world-wide often cause environmental problems associated with their safe disposal. However, the problem could be resolved using edible mushroom species, which possess an appropriate enzymatic mechanism to bioconvert lignocellulosic wastes into value-added products such as food, fodder or plant fertilizer [4, 5].

The fungus *Pleurotus* also called as oyster mushroom belongs to the phylum Basidiomycota, class: Agaricomycetes, family: Pleurotaceae [6]. The oyster mushrooms are the second most important mushrooms cultivated across the world, accounting for 25% of total world production of cultivated mushrooms [7]. Cultivation of the oyster mushroom, *Pleurotus* spp., has increased greatly throughout the world during the last few decades [8, 9]; in 1997 it accounted for 14.2 % of the total world mushroom production. Its popularity has been increasing due to its ease of cultivation, high yield potential and high nutritional value [10]. Although, commonly grown on pasteurized wheat, rice straw and sawdust, it can be cultivated on a wide variety of lignocellulosic substrates, enabling it to play an important role in managing organic wastes [11, 12]. Oyster mushroom species are known to produce lignolytic and hydrolytic enzymes and they can therefore be adapted for growth within a wide variety of lignocellulosic wastes with an increase in the availability of carbohydrates and biomass accumulation [13].

A linear mycelial growth test was used to measure the average mycelium growth in the cultivation of *Lentinula edodes* with sunflower seed halls substrate in combination with different ratios of wheat bran, poplar sawdust or oat straw [14]. The recent study on mycelial linear growth was performed by Lin-Zhi et al. [15] on *Flammulina velutipes*. Thus, reducing the time required for the culture cycle is necessary to decrease production costs. Therefore, methods that increase either mycelium growth or fructification are of interest. The study was undertaken to evaluate the linear growth and yield of four different *Pleurotus* spp. (*Pleurotus ostreatus* white, *Pleurotus ostreatus* grey, *Pleurotus cornucopiae* var. *citrinopileatus* and *Pleurotus salmoneostramineus*) mushroom on easily available sawdust with rice brawn.

MATERIALS AND METHODS

Mushrooms Sampling and Collection

Four different oyster mushrooms namely *Pleurotus ostreatus* (grey oyster), *Pleurotus ostreatus* (white oyster), *Pleurotus cornucopiae* var. *citrinopileatus* (bright yellow oyster) and *Pleurotus salmoneostramineus* (pink oyster) were collected from plant pathology and fungi lab, College of Science, University of Anbar, Iraq. Four different *Pleurotus* spp. were established by tissue culture technique [16]. The mycelia were propagated and maintained on Potato Dextrose agar medium [17] and incubated at 25±1 °C for 6 d.

Spawn Preparation

One kg of healthy grains of wheat (*Triticum vulgare*) was washed thoroughly in tap water and soaked overnight with water till they become soft. One hundred grams each of these grains was taken in 250 ml conical flasks plugged with cotton and sterilized at 15 lbs of pressure for 20 min. After cooling at room temperature, they were inoculated with 10 mm disc of pure cultures. The flasks were incubated at 25±1 °C in dark [18].

Substrates Preparation

Sawdust and rice bran used as a solid substrate for mushroom cultivation. The 89.8% sawdust, 10% rice bran and 0.2% CaCO3 was mixed, added water and keep overnight. The excess water was drained out and the substrates were transferred in to (30 cm x 10 cm) polypropylene bags of one kg. The bags were sterilized for 30 min at 15 lb/sq inch pressure. After cooling the substrate, each different mushrooms spawn was inoculated separately in triplicate. The inoculated mushroom bags were incubated for one month at 25 °C [19].

Linear Mycelial Growth Test (LMG Test)

The linear growth was determined every three days until 30 days. The cumulative growth and periodic growth in 3 days interval were determined according to Saidu et al. [20].

Cropping and Harvesting

As soon as the mycelia growth has covered the whole substrate, the bags were transferred into mushroom shed. The transferred mushroom bags were placed on the racks to expose light intensity of around 2000-3000 lux units for fruiting bodies development. The cultivation room and the substrate were maintained at 80-90% humidity by regularly spraying of water twice a day. Pin heads were formed after 2-4 days and the fruiting bodies were harvested at the right stage and the beds were again maintained for second and third cropping (Figure 1).

Statistical Analysis

Experimental values are given as means. Statistical significance was determined by one variance (One Way) analysis (ANOVA) by using GenStat Discovery Edition computer program version 7 DE3 (VSN International Ltd., UK). Differences at $P < 0.05$ were considered to be significant. The experiments were used three replicates.

ISSN 2348-6201

RESULTS AND DISCUSSION

The effect of solid substrate (Sawdust and rice bran) on mycelial growth of *Pleurotus ostreatus* (grey oyster), *P. ostreatus* (white oyster), *P. cornucopiae* var. *citrinopileatus* (bright yellow oyster) and *P. salmoneostramineus* (pink oyster) is shown in Table 1. Mycelial growth reached its maximum growth on 30th day recorded by visual observation (Fig. 1). The effect of the solid substrate fermentation and mycelial growth varied according to mushroom species. Sawdust and rice brawn has been referred as the most commonly used solid substrate for mushroom production and it is usually offered for the best results in studies on *P. eryngii* [21].

Table 1. Mycelial growth assessment and yield of fruiting bodies on solid substrate

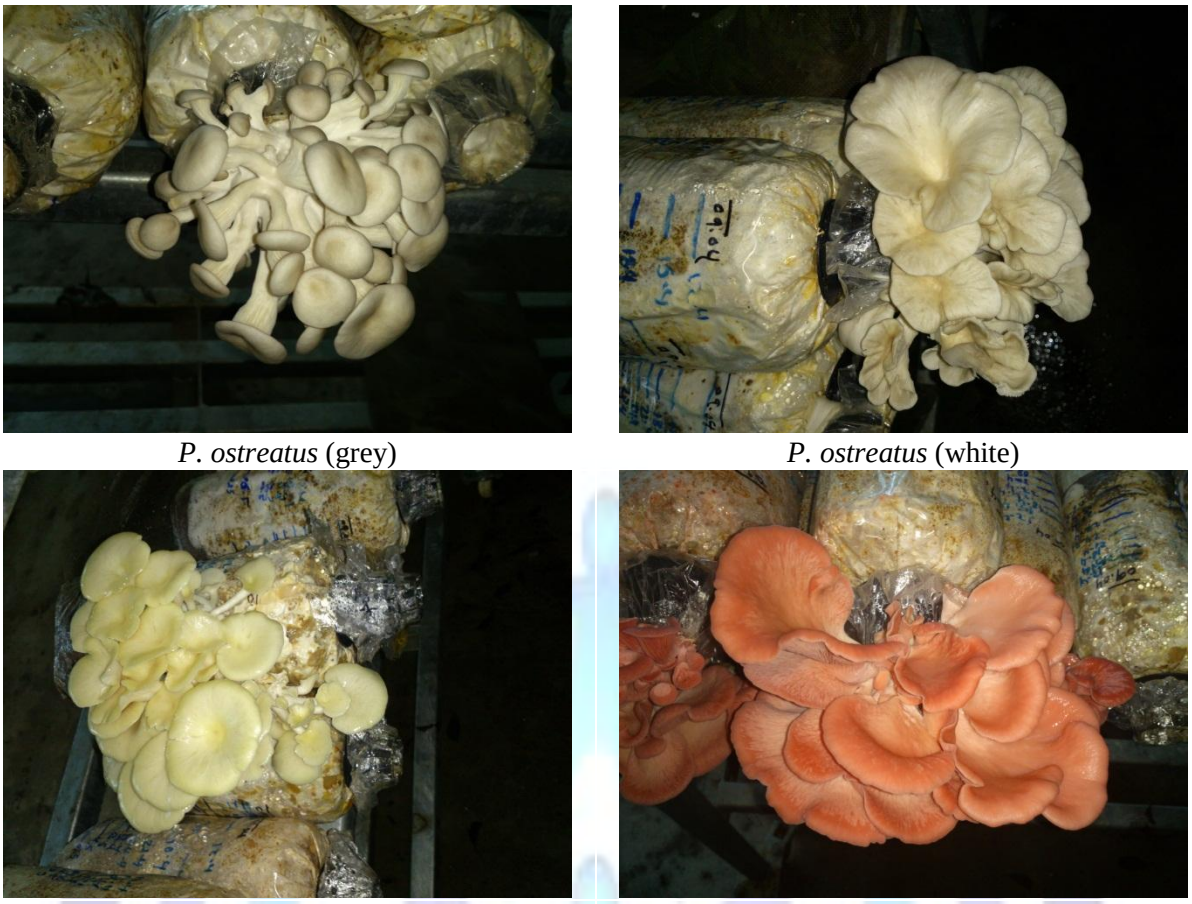
Oyster mushrooms	Average of growth (mm/day)	Cumulative growth in 30 days (cm)	Pin appear date (day)	Fruiting harvested date	Weight of first flush (g)	Average number of fruit bodies	Average weight of fruit body (g)
Grey	1.17	11.74	9.00	11.33	93.8	15.5	7.04
White	1.00	9.99	3.00	9.33	99.2	21.3	4.83
Yellow	0.78	7.82	3.00	10.00	141.0	41.0	3.44
Pink	1.29	12.90	2.00	5.67	96.9	11.0	9.23
LSD P< 0.05	**0.333**	**3.321**	**0.941**	**0.941**	**25.83**	**10.42**	**3.392**

The highest average mycelium growth of 1.29 mm/day followed by 1.17 mm/day was observed in the bags of *P. salmoneostramineus* and *P. ostreatus* grey, respectively. Similarly, the highest cumulative growth of 12.90 cm/day and 11.74 cm/day was recorded in the 30 day old bags of *P. salmoneostramineus* and *P. ostreatus* grey, respectively (Fig. 3 & 4). A recent study recorded the maximum mycelium linear growth of 8.9 mm/day when cultivating king oyster *P. eryngii* on substrate consisting of rice bran, sawdust, soybean straw and rice straw [21].

G: *P. ostreatus* (grey oyster), W: *P. ostreatus* (white oyster), Y: *P. cornucopiae* var. *citrinopileatus* (bright yellow oyster), P: *P. salmoneostramineus* (pink oyster)

Figure 2. Linear mycelial growth of oyster mushrooms in bags (1 kg, wet weight) after 30 days incubation

ISSN 2348-6201



P. ostreatus (grey)

P. ostreatus (white)

P. cornucopiae var. *citrinopileatus* (bright yellow)

P. salmoneostramineus (pink)

Figure 2. Fruiting body emerging on solid substrate

There was a significant increase ($P < 0.05$) of about three fold in the pin head formation of *P. ostreatus* grey (9 days) compared to other mushroom species (2-3 days). Ahmed et al. [22] reported that pin head formation took seven to ten days after the completion of spawn running in *Pleurotus* spp. Obodai et al. [23] reported that pinhead formation took four to six days after the completion of spawn running, with harvest after 10 to 12 days in the case of *P. ostreatus* on different substrate. It is clear that the substrates used in the current study were superior compared to earlier published studies.

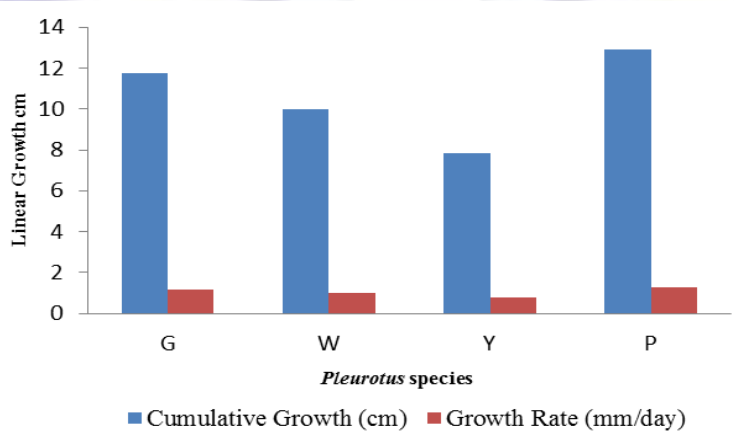


Figure 3. Linear mycelia growth of different oyster mushroom species

ISSN 2348-6201

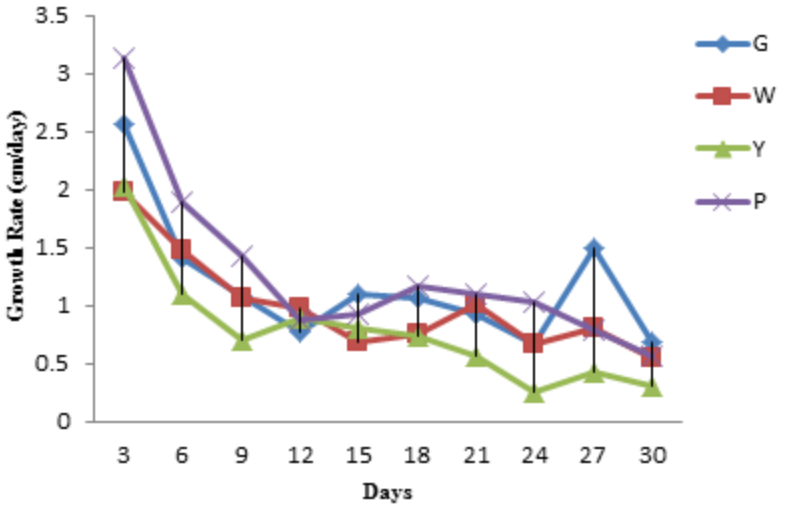


Figure 4. Linear mycelial growth rate of four different oyster mushroom species on solid substrate

Comparing the fruiting body harvesting time of all the four mushroom species, *P. salmoneostramineus* showed a vigorous growth in the fruiting body and the harvesting time required was just 5.67 days. Whereas other species took a longer time (9.33 days for *P. ostreatus* white, 10 days for *P. cornucopiae* var. *citrinopileatus*, and 11.33 days for *P. ostreatus* grey). It is evident from the result that there is no correlation between the rate of mycelia growth and pin appearance. In line with our result, Shukla and Jaitly, [24] have observed similar negative correlation and reported that genetic diversity may play a key role on the mushroom production. Similarly, Kashangura, [25] reported that the primordial initiation and crop production depends on the species and substrate mixture (Table 2).

Table 2. The correlation between characteristics

Oyster mushrooms	Average of growth (mm/day)	Cumulative growth in 30 days (cm)	Pin appear date (day)	Fruiting harvested date	Weight of first flush (g)	Average number of fruit bodies	Average weight of fruit body (g)
Grey	1.17	11.74	9.00	11.33	93.8	15.5	7.04
White	1.00	9.99	3.00	9.33	99.2	21.3	4.83
Yellow	0.78	7.82	3.00	10.00	141.0	41.0	3.44
Pink	1.29	12.90	2.00	5.67	96.9	11.0	9.23
LSD P< 0.05	**0.333**	**3.321**	**0.941**	**0.941**	**25.83**	**10.42**	**3.392**

The yield varied significantly due to the effect of mycelial growth on the substrate. The highest first flush and number of fruiting bodies was achieved in the yellow oyster followed by *P. ostreatus* grey and *P. salmoneostramineus* (Fig. 2 and Table 1).

In conclusion, sawdust and rice bran could be a promising substrate for linear mycelial growth of *P. ostreatus* species. The pin appearance time depends mainly on genetic factors of the mushroom species.

ACKNOWLEDGMENTS

Support for this research by Research Scholarships Programme No. 12/29861 on 12/2012 at foreign universities in Ministry of Higher Education and Scientific Research, Scholarships & Cultural Affairs Directorate, Iraq is gratefully acknowledged to complete this work in MRC, University of Malaya, KL, Malaysia.

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