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Recycling of Lignocellulosic Plant Wastes in Cultivation and Production of Oyster Mushroom (*Pleurotus ostreatus)*

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Abstract: In this research, some local available plant residues, which are including wheat straw, date palm fronds and date palm fibers were used for cultivating and production of oyster mushroom *Pleurotus ostreatus*. The potential for using lignocelluloses wastes as essential substances for substrate preparation in the production of mushroom was used to study their effects on the yield, biological efficiency (BE) were determined. The results showed that the three substrates (wheat straw,date palm fronds, date palm fibers) gave yield of mushroom (143gm, 131gm, 106gm) and BE % gave (76%, 35%, 35%) respectively. The morphological characteristic was determined and influenced by various substrates. The diameter of pileus gave (6.7cm, 6.5cm, 9.75cm) respectively and stipe length gave (5cm, 7cm, 7cm) respectively. The number of effective fruit bodies were(22.5, 4 ,7.5) respectively. In conclusion Mushroom cultivation is one of the efficient ways by which residues can be recycled. *P. ostreatus* grown on different substrates are nutritious with high protein, fiber and low fat. It may also offer economic incentives for agribusiness to examine these residues as valuable resources and develop new enterprises to use them to produce nutritious mushroom products. Therefore, the mushroom cultivation may become one of the most profitable agribusiness that could produce food products from different substrates and help to dispose them in an environment friendly manner.

Keywords: *Oyster mushroom, biological efficiency (BE), morphological characteristic*.

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Introduction

Cultivation of edible mushrooms might be the only current process that result the production of protein-rich food with the reduction of environmental pollution(1).It represents one of the most efficient biotechnological processes for lignocelluloses' organic waste recycling (2).Mushrooms of *Pleurotus* spp. are commonly known as oyster mushrooms that occupy the second position among cultivated edible mushrooms worldwide due to their nutritional and medicinal values. Mushroom cultivation presents an economically important biotechnological industry that has distinguish expanded all over the world in the last few years (3). Mushroom cultivation mainly depends on the agriculture crop residues. These crop resides are abundantly available in our country It is primarily used as cattle feed and remaining part is burnt or

spread in the field. Agro-wastes contain about 80% cellulose, hemi-cellulose, and lignin which are not easily degradable, but most of the edible fungi posses' enzyme system which degrade these components. Thus mushroom cultivation can avoid environmental pollution by recycling agricultural wastes and can also convert straw into easily digestible animal feed (4).Oyster mushroom has been widely cultivated in many different parts of the world. It has abilities to grow at a wide range of temperatures utilizing various lignocelluloses (5).Oyster is the most common cultivated variety due to its simplicity and low cost cultivation technology(6).It has the ability to degrade residues (primary decomposers) from agricultural wastes and convert them into protein rich biomass (7).It is one of the most second important edible mushrooms after *Agaricus bisporus* for production and consumption worldwide and has broad adoptabilityamong the mushroom growers reasons being 1) to be grow it on various organic wastes ofagricultural, forest and industrial origin under wide range of agro climatic conditions from30-10 ℃ with 60- 90 percent humidity; 2) easy to grow, easy to manage in crop room andeasy to post harvest; 3) it is nutritionally and medicinally rich with good taste Large volumes of rice and wheat straw are produced as agricultural by-products (8). Currently, the agricultural wastes are disposed of through open-field burning, which leads to serious environment pollution problem. If they can support the growth of oyster mushroom, then it would be one of the solutions to transform these inedible wastes into an accepted edible biomass of high market value, and serve as a cheap source of substrate for mushroom grower(9).However, the utilization of rice straw and/or wheat straw in oyster mushroom cultivation is not popular in China for its low yield and low biological efficiency (10), (11).Oyster is the most common cultivated variety due to its simplicity and low cost cultivation technology. It has the ability to degrade residues (primary decomposers) from agricultural wastes and convert them into protein rich biomass (12).

The aim of this study is to evaluate the better performance of oyster mushroom in different substrate compositions as well as to find out the better substrate for mushroom cultivation and preservation of the environment from environmental pollution.

Materials and methods

Microorganism and spawn preparation

Pleurotus ostreatus, strain obtained from Food Biotechnology Research Center of Cairo university, was grown on the potato dextrose agar (PDA) medium at 25 ℃ for regular subculture.The cultures were later authenticated by microscopic and macroscopic features Oyster mushroom spawn was prepared in 250-ml glass bottles filled with wet wheat grains and then autoclave sterilized at 121 ℃ for 80 min. After cooling down to room temperature, the sterilized wheat grains bottles were inoculated with 1 cm^2 mycelial agar discs. The spawn was incubated in the laboratory at 26 ± 2 ℃ and 70% relative humidity for two weeks.

Substrate preparation, inoculation, and incubation

Three types of Substrates were cultivated (wheat straw, datepalm fronds, date palm scaly fibers). These substrates were completely dried under the sun, and chopped into 4 cm lengths, and soaked in clean tap water for overnight before substrate preparation.

After draining the excess water, they were used as substrate. The substrate cultivation was filled into polyethylene bags and autoclave sterilized at 121 °C for 80 min. The substrates were cooled down to room temperature, they were inoculated with 20 g of oyster mushroom spawn. using layers in depth 5 cm, spawn added then another layer of substrate added until filling then closed. Twenty replicate polyethylene bags were used for each substrate. Incubation was achieved darkly at 25°C ± 1°C for 3 weeks until the complete growth of mycelia in the whole packet. 10°C for 48 h was carried out as cold shock to induce pin heads formation, packets opened with 25°C ± 1°C, 80-90% relative humidity, lighted using fluorescent light, and aerated twice per today so primordial form that developed to fruiting bodes, which harvested in best yield size (12). The spawn run period to total colonization (the number of days from inoculation to complete colonization of the substrate by the mycelium) was recorded.

Yield Performance, harvest and determination of biological efficiency

After the primordia appeared on the top layer of substrate in each polyethylene bag, then Mushrooms were harvested when the mushroom cap surface were flat to slightly up-rolled at the cap margins. The harvested fruiting bodies in each bag were then counted and weighed. At the end of the harvest period, these data were used to calculate the biological efficiency and mushroom weight.

Mushroom weight (g) = Total weight of harvested fresh mushrooms per bag/total number of mushroom.

The developed fruiting bodies counted to determine the number of effective ones; tiny and deformed fruiting bodies were discarded at the time of counting. To obtain the average weight of individual fruiting bodies, the weight of each flush was taken and divided by the number of fruiting bodies. Average length, breadth of ten randomly selected pili (cap) were measured (mm) using slide calipers. The length of the stipe was also measured (mm). Fruiting bodies were collected as cluster form from spawn bags. The whole cluster of fruiting bodies was considered for biological yield measurement, and the cleaned fruiting bodies (separated from cluster by removing lower tough and dirty portion) were weighted to obtain economic yield (13). The biological efficiency was determined to obtain the growth potential of the fungal strain. The conversion percentage (biological efficiency) was determined (14) as follows: biological efficiency (BE) = (grams of fresh weight of mushroom/dry weight of substrate) × 100%.

Results and Discussions

This study aimed to evaluate the growth and yield of oyster mushroom (*Pleurotus ostreatus*)on three different substrates : wheat straw, fronds of date palm, and scaled fibers of date palm. The result demonstrated significant differences in both micelial growth and mushroom yield, which can have attributed to the inherent nutritional composition and physical properties of the substrates. A substrate is any material that serves a medium of growth for a living thing in which enzymes can act upon and break it to release nutrients for growing organisms. There are a range of waste that can be used for oyster mushroom cultivation, but it depends on the basis of availability of the substrate and its cost. The important of good substrate is an important requirement for the better growth and higher yield of mushroom (15).The remarkable ability of edible fungi to use

various substrate and transform complex organic compounds into simplex by-products can be put to use to convert and utilize the vast agricultural and green waste into thevaluable and usable form (16).

1. Mycelial growth and primordial initiation

Three different types of substrates were investigated to determine the growth and yield of *P. ostreatus*. Myceliagrowth and spawn production are the preliminary step that creates suitable internal conditions for fruiting. Thus, outstanding growth of mycelium is a vital factor in mushroom cultivation (17).Colonization of the substrate as shown in Fig.(1) was completed in between 22.40-26.00 days of incubation. and that indicating the time required for mycelium to fully colonize the substrates. Similarly, primordial initiation on various substrates was also observed in between 26.40 to 31.60 days of incubation. The total day for the first harvest of mushroom took between 32-37 days, depending on substrate used. These results are agreed with previous studies (18) which highlight the importance of adequate mycelia growth for successful fruiting. It is noteworthy that substrates with higher cellulose and lignin content, such as wheat straw, tended to facilitate faster mycelia colonization, which may explain the faster initiation observed on this substrate (19).

2. Biolodical yield and yield conformance

The Biological yield, defined as the total fresh weight of mushroom harvested showed considerate variability across substrates. Biological yield varied significantly due to effect of different substrates. The results showed in Fig. (2) that maximum biological yield was demonstrated on the wheat straw was the best in the yield performance it gave average weight of fruit bodies 143g followed by 131g in front of date palm and 106 g scaled fibers of date palm. The result demonstrates that the best incubation time for production was between 40-50 days in both substrate (wheat straw), this aligns with finding from a study demonstrated by (20) who observed similar trends in yield performance when uses eight lignocellulosic by-products as substrates for cultivation of the Oyster mushroom, *Pleurotus ostreatus*. Wheat straw, a highly lignocellulosic material, provided optimal condition for *P. ostreatus*. Mycelial growth contributing to higher yields. The results also showed that incubation times between 40-50-days were optimal for mushroom production, as both Wheat straw and fronds of date palm showed peak yields within this timeframe which agreed with (20), (21].

3. Number of effective fruiting body

Effective fruiting body is the edible part of mushroom on different substrates was shown in Fig. (3). There was significant variability on different substrates under this study The number of effective fruiting body with average 23 of fruit bodies and the maximum number of effective fruiting body was recorded on wheat straw. The minimum number of effective fruiting body as shown in Fig. (4) was recorded on fronds of date which gave 4 and in scaled fibers of date palmgave average of 7.5 effective fruit bodies. This variability can be attributed to the chemical composition of the substrates Other studies was reported by (22). Higher number of effective fruiting body might happen due to the presence of glucose, fructose and trehalose in the substrate, reported by (23].

4. Stalk length:

The length of stalk, is an important factor in the economic yield of mushroom, varied significantly among substrates.Length of the stalk of *P.*

ostreatus was differed on different substrates at 1% level of significance. In case of 1st flush highest stalk length was recorded on fronds an in scaled fibers of date palmFig.(4) that gave average of 7cm. while wheat straw show shorter stalk(5 cm.) The lowest stalk length was recorded on wheat straw that gave 5cm which is important in economic yield but does not always correlate with yield.

5. Pileus diameter

The Pileus diameter, is another key characteristic of *(Pleurotus ostreatus*) fruiting bodies, varied across substrates. Diameter of pileus differed on different substrates. In case of 1st flush pileus diameter was found the largest pileus diameter (9.5cm) on scald fibers of date palm and in wheat straw gave average of 6.7cm in diameter and with fronds of date palm was 6.5 cm. there is no difference significant in pileus diameter between fronds of date palm and wheat straw as the increases of pileus diameter the yield decreases, In this study , scaled fibers of date palm showed larger pileus diameter , but yielded lower mushroom , which support the notion that increased fruit body size may lead to decreased biological efficiency as shown in Fig. (4).

6. Biological Efficiency (BE)

The highest biological efficiency was calculated in treatment with straw of wheat, it gave average 76% while with fronds of date palm gave (35%) and lowest in dead scaled fibers gave 30%The biological efficiency for different substrates ranged from 30 to 76% and its differs due to combined supplements with basal ingredient that results better mushroom quality as well as. the biological efficiency was proportional with the yield percent as shown from figures (2) and (3). the highest yield was in wheat straw substrate the lowest 106gm yield with yield percent and BE 35 % gave average about 30%. And this result agrees with (24) in his study on different type of agricultural waste plants.The differences in BE emphasize the importance of substrate choice in optimizing both yield and resource utilization. Wheat straw gave high BE due to its high lignocellulosic content, which support both mycelial growth and fruiting bodies development more efficiency than other substances. The lower BE % with date palm substrates could be due to their lower nutritional content, which may limit the mycelium ability to fully colonize and produce fruiting bodies (25].

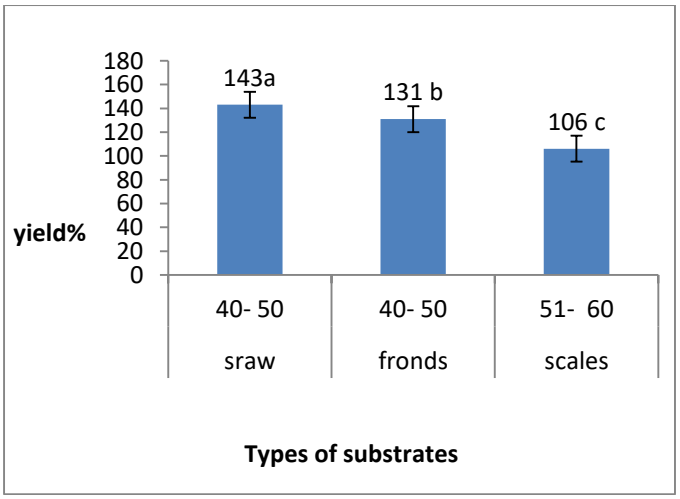

Figure (1): Effect of incubation period on biological yield of oyster mushroom grown on different substrates

Mean mushroom weight (g), cap diameter (cm), stipe length (cm) of oyster mushroom (*Pleurotus ostreatus*) grown on different substrates. Letters indicate means among substrate followed by the same letter are not statistically different ($P > 0.05$).

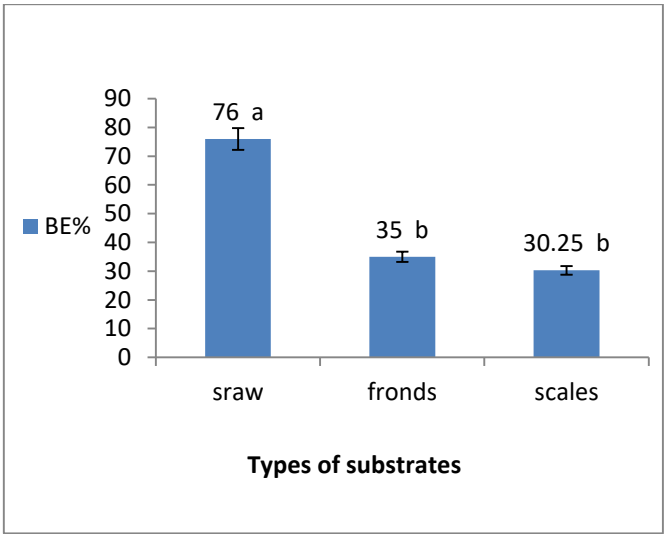

Figure (2): Effect of different substrates on biological efficency (BE%) of oyster mushroom

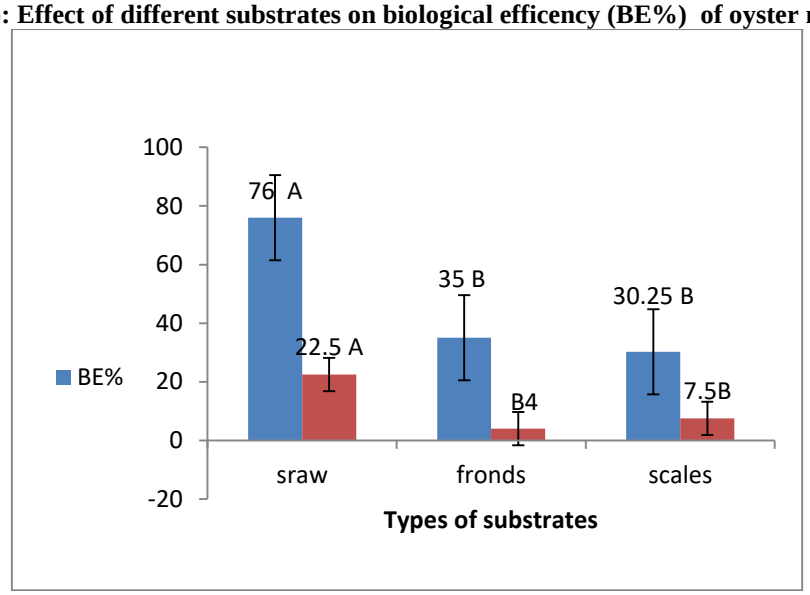

Figure (3): Effect of substrates on BE % and No. of fruit bodies of oyster grown on different substrate

The results in (Fig. 4) show the variation morphological characters which include the stipe length and mushroom cap diameter was observed in the three substrates used in this study. They showed high number of fruit bodies when grow on straw and in fronds gave less productively of fruit bodies Relatively smaller mushroom cap diameter and longer mushroom stipe length are undesirable characteristics as for marketable quality. Environmental conditions as well as supplementation of substrates with various additives including nitrogen sources have been reported to improve growth, yield and quality of mushrooms (26). The statistical analysis showed significance differences in number of effective fruiting bodies when grown on wheat straw.While in scales of date

palm showed significance differences in pileus diameter.

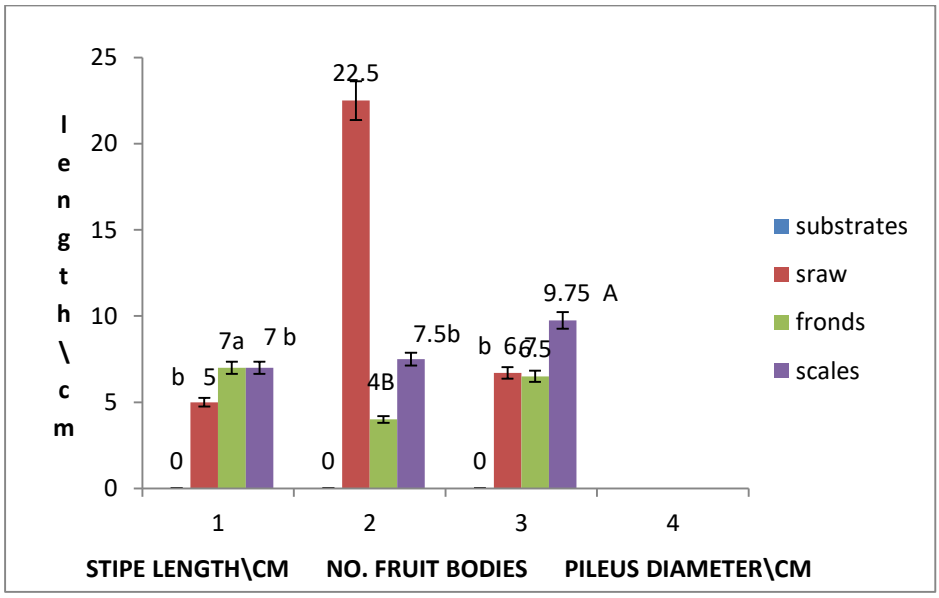

Figure (4): The morphological characters of oyster mushroom (*Pleurotus ostreatus*) grown on different substrates.

Mean mushroom fruit bodies, cap diameter (cm), stipe length (cm) of oyster mushroom (*Pleurotus ostreatus*) grown on different substrates. Letters indicate means among substrate followed by the same letter are not statistically different ($P > 0.05$).

Statistical analysis

Descriptive statistics: Before performing analysis of variance (ANOVA). it is helpful to compute Descriptive statistics (mean , standard deviation and rang) for each substrate to sumerize the data. Since the study compares the performance of oyster mushroom (*Pleurotus ostreatus*)on three different substrates (wheat straw , fronds of date palm and scaled of date palm). The analysis of one way ANOVA by using Least Significance Differences (LSD) was used to compare the means of different variables across the three substrates at level 0.05 level.

The varables for ANOVA were :

1.Mycelial growth (days to complete colonization

2.Time to primodial initiation

3.Time to the first harvest

4.Biological efficiency

5.Number of fruit bodies

Conclusion

From the results, wheat straw showed as the most promising substrate for the cultivation of *Pleurotus ostreatus*, yielding the highest biological yield, number of fruiting bodies and biological efficiency. On the other hand, while fronds and dead scales of date palm showed potential, their lower yield and biological efficiency indicate that they may not be as suitable for large scale mushroom production.Mushroom cultivation is one of the efficient ways by which residues can be recycled. *P. ostreatus* grown on different substrates are nutritious with high protein, fiber and low fat. It may also offer economic incentives for agribusiness to examine these residues as valuable resources and develop new enterprises to use them to produce nutritious mushroom products. Therefore, the mushroom cultivation may become one of the most profitable

agribusiness that could produce food products from different substrates and help to dispose them in an environment friendly manner.

Recommendation

Mushroom growers need both experience and education. Mushrooms are often thought of as an easy crop to produce with a high price potential. People often find mushroom cultivation is not as easy as they thought it would be. Much of the necessary knowledge must be acquired through practical experience, but understanding the principles of mushroom cultivation demystifies the process, allowing the grower to successfully adapt and develop cultivation methods.

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