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Effect of Different Agro-Industrial Wastes on the Growth and Yield of Edible Mushrooms (*Pleurotus florida*)

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ABSTRACT

The study carried out investigate the effect of different agro-industrial wastes and mixed substrates with rice straw (w/w) on caltivation of oyster mushroom (*Pleurotus florida*). Mixed substrates with rice straw at rate 1:1 by weight had valuable effect on the spawn running, duration pinhead's formation and fruiting bodies formation. Results regarding to the time required for completion were found within the period ranged from 11.25 - 27, from 23.5 - 51 and from 27 - 54 days respectively. When mixed substrates with rice straw the spawn running, pinheads and fruiting bodies formation were found within the period ranged from 10.5 - 19.25, from 22 - 26.5 and from 27 - 32 days respectively. Mixed substrates with rice straw reduce the spawn running period of *P. florida* over control substrates. Therefore, reduce pinheads and fruiting bodies formation period. The highest total weight of mushroom harvested per 1 kg dry mixed substrate was recorded on rice straw with waste paper (RS + WP) ~ 853.87 g and gave the maximum biological efficiency 85.49 %. Mixed substrates with rice straw increase in biological efficiency of *P. florida* over control substrates.

Keywords **:** Agro-industrial wastes; Mixed substrates; *Pleurotus florida.*

INTRODUCTION

Cultivation of edible mushrooms is a biotechnological process for lignocellulosic organic waste recycling. It might be the only current process that combines the production of protein-rich food with the reduction of environmental pollution (Beetz and Kustudia, 2004). Since cultivated mushrooms can grow on agricultural and industrial wastes they constitute a source for obtaining food protein from such wastes and thus they can be marshaled to aid in solving many problems of global importance including protein shortages, resources recovery and environmental management. Different plant waste materials were used as substrate for cultivation of mushroom (Zadrazil, 1980). The wheat straw and combination of it with cotton seed cake and rice bran have been found more capable for substrates to grow mushroom on commercial point of view by farmer's community as a cheap source of protein (Khan, *et al.*, 2019).

Mushrooms have the ability to degrade several lignocellulosic substrates by produced natural materials from agro- industrial wastes (Barny, 2009).

In this connection, *Pleurotus florida* is a typical edible species of oyster mushroom. This fungus is widely distributed in Africa and other tropical countries. It belongs to phylum basidiomycetes, order agaricales and family tricholomataceae (Zoberi, 1972, Alexopolous *et al*., 1996, Alofe *et al*., 1998).

P. florida is an excellent edible and highly nutritious mushroom which is a common species in Egypt, tropical West Africa and Southern part of Asia. The edible fruit bodies develop in large numbers as a group on fallen trees, logs of wood and wooden poles. The cap measurement may range from 1.5 to 7.5cm in diameter while the stipe is 0.5 to 2.5cm in length, annuls is absent and the spore print is cream – white in colour (Jonathan, 2002).

P. florida is suitable for bioremediation of contaminated soils because of its ability to degrade highly condensed polycyclic aromatic hydrocarbons (PAH) and its high tolerance of these substrates (Adenipekun and Gbolagade 2006). Generally, the nutritive value of agro by-product was improved upon fungal treatment (Nasehi, *et al.*, 2017). Tolulope, *et al*., (2021) reported that medicinal leaves could be used to increase the yield of *P. florida* and gave a better yield with rice straw.

The objective of the study was to determine the effect of mixed substrates with rice straw (w/w) on cultivation of oyster mushroom (*Pleurotus florida*) on different agro-industrial wastes in order to produce protein-rich food with protection of environment from pollution.

MATERIALS AND METHODS

***Pleurotus florida*:**

The sorghum spawns of *P. florida* were obtained from Mushroom Unit, National Research Center (NRC), Douki, Cairo, Egypt.

Culture and maintenance medium:

The *Pleurotus florida* was grown on malt agar medium. (25% malt extract and 1.5% agar (Oxoid) at 28 ^{o}C). The fungal culture was maintained at 5^{o}C and subcultured monthly (Atlas, 1995) until used.

Substrate preparation and cultivation condition:

All substrates were obtained from Mansoura city, Dakhlia, Egypt. Rice straw (*Oryza sativa*), sawdust, waste paper of Handmade paper industries and cardboard industries were used as a base substrate. These substrates were chopped

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into small pieces of 2 – 5 cm. Rice straw (RS) and Sawdust (SD) were soaked in water for 24 hours before use. Waste paper (WP) and Cardboard industrial (CI) were soaked in hot water for 3 hours to remove dyes before use (Espindola *et al*., 2007). All substrate was boiled for 15 – 20 minutes and then spread over clean. surface in thin layers for cooling and draining of the excessive water. Rice straw (RS), Sawdust (SD), Waste paper (WP), Cardborad industries (CI) are mixed with rice straw at rate 1:1 by weight {(RS + SD), (RS + WP), and (RS + CI)}.

Plastic bag was used in production experiments. Substrates were spawned at a ratio 4% of sorghum spawn (w/w). Each bag was closed with a plastic neck. The spawned bags were then incubated at 25 – 30 ºC and 60 – 65% relative humidity for 2 – 3 weeks in a well-ventilated, semi dark room until spawn run was completed (El-Sawah, 2000 & Lawal, *et al.,* 2011).

Cropping:

After a complete spawn run, the bags were opened after 2 weeks in case with waste paper and cardboard, 3 weeks for rice straw and 4 weeks for sawdust, when the mycelium had completely covered the substrate. The compact mass of the substrate and mycelium was watered daily with distilled sterilized water from the second day of opening the bags. Within 3 – 8 days of opening, pin head fruiting bodies (4 – 5 cm in diameter) appeared on all sides of the bag. Young mushrooms attained the normal size in about 3 – 5 days when the first crop was harvested from each bag. Mature fruiting bodies were harvested at different periods and the fresh weight recorded immediately after the harvest. The time taken for the spawn running and appearance of pin heads was also recorded. Biological efficiency (BE) was calculated as percentage yield of fresh mushroom fruiting bodies in relation to dry weight of the substrate.

Statistical analysis:

Data was carried out according to SPSS program 10 for Windows (SPSS, 1999) using LSD test to compare between mean values of all treatments.

RESULTS AND DISCUSSION

Growth of Pleurotus *florida* on different substrates:

Table (1) show, the growth of oyster mushroom (Pleurotus *florida)* on tested different agricultural wastes, namely rice straw (RS), sawdust (SD), waste paper (WP) and cardboard industrial (CI).

Spawn running:

The spawn running, pinheads and fruiting bodies formation are three important phases in the cultivation of oyster mushroom, require proper humidity and temperature. Temperature 25 ºC for spawn running and 17 – 20 ºC for fructification showed good results (Shah *et al*., 2004). The shortest spawn running period was determined being 11.25 days on waste paper and the longest was 27.00 days on sawdust as shown in Table 1. While Lalithadevy and Many (2014) documented the completion of spawn running on paddy straw to between 16 and 25 days.

Pinheads formation:

Small pinheads like structures were observed, (Shah *et al*., 2004). The pinheads of *P. florida* started appearing 3.00 – 38.50 days after spawn running, rice straw pinned in 23.50 days followed by sawdust pinned in 32.75 days, waste paper pinned in 45.75 days, cardboard took a longer time to pinned 51.00 days as shown in Table 1.

These results are in agreement with those reported by Kulshreshtha *et al*., (2010) in their bioremediation of industrial waste through mushroom cultivation.

Fruiting bodies formation:

Agro-industrial wastes tested were different in suitability for oyster mushroom (*Pleurotus* spp.) cultivation. The agro-industrial wastes had valuable effects on fruiting bodies formation. The fruiting bodies of *P. florida* appeared 3.00 – 5.00 days after pinheads formation and took 27.00 – 54.00 days later after spawning. Cardboard took a shorter time to fruiting bodies formatted in 3.00 days after pinheads formation followed by rice straw fruiting bodies formatted in 3.50 days. Sawdust and waste paper took a longer time to fruiting bodies formatted in 4.25 and 5.00 days after pinheads formation, while cardboard took a longer time to fruiting bodies formatted was determined as average of mycelial formation (MF) being 39.16 days later after spawning (Table 1).

These results are in agreement with those reported by Mandeel *et al*., (2005) and Kulshreshtha *et al*., (2010) in their cultivation of oyster mushroom (*Pleurotus* spp.) on various lignocellulosic wastes and bioremediation of industrial waste.

Table 1. Spawn running, pinheads formation and fruiting bodies formation of *P. florida* at 25 – 30 ºC on different substrates by days completion.

Substrates	Spawn running (days)	Pinheads formation (days)	Fruiting bodies formation (days)	MF
Rice straw	20.50	23.50	27.00	23.66
Sawdust	27.00	32.75	37.00	32.25
Waste paper	11.25	45.25	50.25	35.58
Cardboard industrial	12.50	51.00	54.00	39.16

MF= average of mycelial formation

Effect of mixed substrates with rice straw (w/w) on *P. florida* growth :

Mixed substrates with rice straw at rate 1:1 by weight had valuable effect on the spawn running, duration pinheads formation and fruiting bodies formation. The obtained results are recorded in Table 2

The time of spawn running of *P. florida* was determined as average 10.50 – 19.25 days. The shortest spawn running period was determined as average 10.50 days on rice straw with waste paper (RS + WP) and rice straw with cardboard industrial (RS + CI). The longest was 19.25 days on rice straw with sawdust (RS + SD) as shown in Table 2.

The pinheads started appearing 2.75 – 16.00 days after spawn running. The shortest pinheads formation period was determined as average 22.00 days on rice straw with sawdust (RS + SD) and the longest period was 26.50 days on rice straw with waste paper (RS + WP) and rice straw with cardboard industrial (RS + CI).

Table 2 show the *P. florida* growth parameter. The fruiting bodies appeared 3.50 – 5.50 days after pinheads formation and took 27.00 – 32.00 days later after spawning. The shortest fruiting bodies formation as average of mycelial formation (MF) period 22.08 days on rice straw with sawdust (RS + SD) and the longest as average of mycelial formation (MF) period was 23.00 days on rice straw with waste paper (RS + WP).

Mixed substrates with rice straw reduce the spawn running period of *P. florida* over control substrates (Fig. 1). and consequently reduced pinheads and fruiting bodies formation period .

These results are in assurance with those obtained by Akyuz and Yildiz (2008) and Kulshreshtha *et al.*, (2010) in their determinatin of cellulosic wastes for the cultivation of *Pleurotus eryngii* (DC. ex Fr.) Quel and bioremediation of industrial waste through mushroom cultivation. Tolulope, *et al*., (2021) reported that medicinal leaves could be used to increase the yield of *P. florida.*

Table 2. Effect of mixed substrates with rice straw on growth (days) of *P. florida* at 25 – 30 ºC .

Substrates (w/w)	Spawn running	Pinheads formation	Fruiting bodies formation	MF
Rice straw + Sawdust	19.25	22.00	27.00	22.08
Rice straw + Waste paper	10.50	26.50	32.00	23.00
Rice straw + Cardboard industrial	10.50	26.50	30.00	22.30

MF= average of mycelial formation

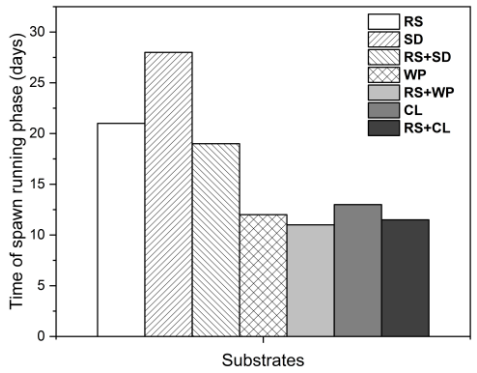

Fig. 1. Effect of mixed substrates with rice straw on growth of *P. florida.*

RS ; Rice straw. SD; Sawdust. WP; Waste paper. CI; Cardboard industrial. RS + SD ; Rice straw + Sawdust. RS + WP ; Rice straw + Waste paper . RS + CI ; Rice straw + Cardboard industrial .

Yield and biological efficiency :

Biological efficiency was calculated (as the percentage) because some substrates were denser than others (Akyuz and Yildiz, 2008).

The analysis of variance different substrate effects on yield of *P. florida* revealed significant differences ($P<0.05$) between used substrates. Rice straw was superior to all the other substrates. Tested agro-industrial wastes recorded various flushes. The first flush of crop gave 50% of the yield obtained in all the by-product substrates tested. The yield of *P. florida* and biological efficiency was shown in (Table 3). The highest total weight of mushroom harvested per 1 kg dry substrate was of rice straw was 593.28 g , followed by sawdust 204.41 g , and the lowest yield was recorded on waste paper.

Table 3. Effect of different substrates on Yields and Biological efficiency of *P. florida*.

Substrates	Total fresh weight of fungus g/kg substrate dry weight	Biological efficiency (%)
Rice straw	593.28	59.33
Sawdust	204.41	20.44
Waste paper	46.50	4.65
Cardboard industrial	170.21	17.02
LSD at 5 %	4.28	0.428

The biological efficiency of *P. florida* production varied in different used substrates (Table 3). The maximum biological efficiency of was recorded with rice straw (59.33 %), followed by sawdust (20.44%).

Effect of mixed substrates with rice straw (w/w) on yield and biological efficiency :

Rice straw is the principal substrate for oyster mushroom growing, although adequate production can be achieved through use of wheat straw with the addition of supplements that substantially increase the yield per unit weight (Baysal *et al*., 2003).

The analyses of variance mixed substrate effects on yield of *P. florida* revealed significant differences ($P<0.05$) between used substrates. Rice straw was superior to all the other substrates.

The effect of mixed substrates on yield performance of *P. florida* was shown (Table 4). The highest total weight of mushroom harvested per 1 kg dry mixed substrate was recorded on rice straw with waste paper (RS + WP) ~ 853.87 g , followed by rice straw with cardboard industrial (RS + CI) ~ 565.45 g , and the lowest yield was recorded on rice straw with sawdust (RS + SD) ~ 398.80 g.

Results of the biological efficiency of *P. florida* production on mixed substrates with rice straw in Table 4 show that the maximum biological efficiency of 85.49% was recorded on rice straw with waste paper (RS + WP), followed by rice straw with cardboard industrial (RS + CI) 56.55%.

Mixed substrates with rice straw increased in the biological efficiency of *P. florida* over control substrates (Fig. 2).

The results are in agreement with those reported by Akyuz and Yildiz (2008) and Kulshreshtha *et al.,* (2010) in their evaluation of cellulosic wastes for the cultivation of *Pleurotus eryngii* (DC. ex Fr.) Quel and bioremediation of industrial waste through mushroom cultivation.

Table 4. Effect of mixed different substrates with rice straw on Yields and Biological efficiency of *P. florida.*

Substrates (w/w)	Total fresh weight of fungus g/kg substrate dry weight	Biological efficiency (%)
Rice straw + Sawdust	398.80	39.88
Rice straw + Waste paper	853.87	85.49
Rice straw + Cardboard industrial	565.45	56.55
LSD at 5 %	2.45	0.245

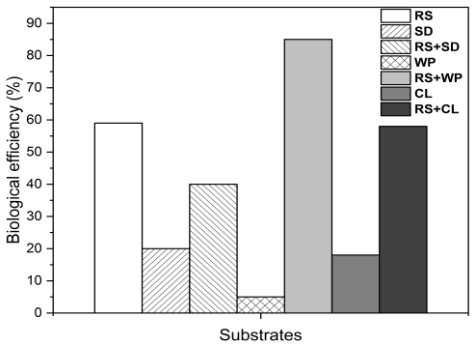

Fig. 2. Effect of mixed substrates with rice straw on Biological efficiency of *P. florida.*

RS ; Rice straw. SD; Sawdust. WP; Waste paper. CI; Cardboard industrial. RS + SD ; Rice straw + Sawdust. RS + WP ; Rice straw + Waste paper . RS + CI ; Rice straw + Cardboard industrial .

Tolulope, *et al.*, (2021) reported that cultivation of *P.florida* on agro-wastes can be an economical and harmless method of waste disposal. Despite the differences in the yield and biochemical composition of the mushrooms; the over all nutritional potentials of the mushrooms were good on all the lignocellulosic wastes.

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تأثير المخلفات الزراعية والصناعية المختلفة على زراعة فطر عيش غراب (*Pleurotus florida*)

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تهدف هذه الدراسة معرفة تأثير المخلفات الزراعية والصناعية المختلفة ، وكذا خلط هذه المواد مع قش الأرز لاستخدامها فى زراعة فطر عيش غراب الأويستر نوع بليروتس فلوريدا *Pleurotus florida* ، فعند خلط المواد المستخدمة فى الزراعة مع قش الأرز بنسبة (١ : ١) وجد لها تأثير ملحوظ على مراحل نمو وانتاج الفطر . ووجد ان بالزراعة على المخلفات الزراعية والصناعية المختلفة ان الوقت اللازم لنمو الاسبون وتكوين رؤوس الدبابيس وتكوين الاجسام الثمرية تم خلال الفترة من ١١,٢٥ إلى ٢٧ ومن ٢٣,٥ إلى ٥١ و من ٢٧ إلى ٥٤ يوم على التوالى . وعند خلط هذه المخلفات مع قش الأرز كانت النتائج المتحصل عليها للوقت اللازم لنمو الاسبون وتكوين رؤوس الدبابيس وتكوين الاجسام الثمرية فى خلال الفترة من ١٠,٥ إلى ١٩,٢٥ و من ٢٢إلى ٢٦,٥ و من ٢٧ إلى ٣٢ يوم على التوالى .ويتضح من ذلك ان خلط المواد المستخدمة فى الزراعة مع قش الأرز أدى إلى تقليل فترة نمو الاسبون وبالتالى تقل فترات تكوين رؤوس الدبابيس وتكوين الاجسام الثمرية للفطر. كما تم الحصول على أعلى محصول للفطر عند خلط قش الأرز مع مخلفات الورق حيث تم الحصول على ٨٥٣,٨٧ جم / كجم مواد مختلطة . كما سجل خليط قش الأرز مع مخلفات الورق أعلى كفاءة بيولوجية للفطر وكانت ٨٥,٤٩ % أى ان : خلط المواد المستخدمة فى الزراعة مع قش الأرز أدى إلى زيادة الكفاءة البيولوجية للفطر .