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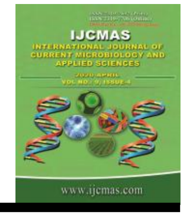

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Effect of Different Agro-wastes Substrates on the Growth and Productivity of the Oyster Mushroom in Upper Egypt

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A B S T R A C T

The Oyster mushroom (*Pleurotus cystidiosus*) was cultivated on different agro-wastes as substrates such as; sugarcane bagasse (ScB), rice straw (RS), and wheat straw (WS), whereas the fourth formula composed of combination of these three substrates in a proportion of 1: 1. The ScB, RS and WS agro-wastes substrates were selected as the most favorable lignocellulosic sources. *P. cystidiosus* can efficiently degrade the agro-wastes and grow at wide range of temperatures. The effects of growing the Oyster mushroom on different lignocllulosic agro-wastes as substrates were tested through evaluating the levels of various parameters in the growing mycelium including: moisture contents, yield, total carbohydrates, crude fat, crude protein, crude fiber, ash, and minerals contents. The WS showed the highest yield (97% Bio efficiency), maximum crude protein (23%), and phosphorus content of (890 mg/ 100 g of dry mushroom). The ScB recorded minimum moisture (88%) and crude fiber content (9%). However, the maximum moisture (91%) and highest fruiting bodies size (10 cm) are recorded on RS cultivation. The combination of substrates formula ScB+ WS+ RS expressed the highest carbohydrate content (58 %), ash (7.1%), phosphorus (585 mg/ 100g of dry mushroom), and calcium (320 mg/ 100g of dry mushroom) content. Finally, we concluded that the *P. cystidiosus* of Upper Egypt can grow in a wide range of temperatures and substrates mainly on WS, then Rs, and ScB. The aim of the current work was to study the influences of the different agro-wastes substrates and some nutritional conditions on the mycelial growth of *P. cystidiosus*, during winter season in the Upper Egypt.

Introduction

Recently, the problem of wastes accumulation and the challenges of food production are the most important problems facing the world, especially with the interest of growing and environmental awareness. There is a great interest towards biodegradation of the agricultural wastes in the developing countries such as; rice straw, wheat straw, and sugar cane bagasse). In addition, all the cellulosic residues of the fields are currently used along with the other food wastes to increase the development and production of mushrooms. The Oyster mushroom (*P. cystidiosus*) is the second widely cultivated mushroom worldwide following the *Agaricus bisporus* (Kues and Liu, 2000). *P. cystidiosus* is popular and widely cultivated mostly in the Upper Egypt. The conditions of the Upper Egypt region mainly high temperature are suitable for *P. cystidiosus* growth, in addition to low cost of production technology and the high biological efficiency (BE) (Mane *et al.*, 2007). *P. cystidiosus* belongs to the family Tricholomataceae, and composed of about fourty species that are cultivated in different parts of Egypt. It can grow well on different types of lignocellulosic substrates and at different temperature conditions. Chang *et al.*, (1989) reported that *P. cystidiosus* is ideally suited for cultivation throughout the year in various regions of the tropical countries, and is ranked next to *A. bisporus*, throughout the world in terms of yield and production. Jandiak *et al.*, (1995) reported that *P. cystidiosus* is an efficient lignin degrader which can grow on a wide variety of agricultural wastes, and at a wide range of temperatures. This Oyster mushroom has high economical and medicinal values, due to its chemical composition and\or nutrition. Li *et al.*, (2007) added that several species of Oyster mushroom are very important in the field of medicine acting as strong antioxidants. *P. cystidiosus* possesses antitumor activity, and has hypoglycaemic accents in experimentally induced diabetics (Chorvathova *et al.*, 1993). Oyster mushroom is able to colonize and degrade a large variety of lingocellulosic materials and other wastes, which are produced in agricultural, forest and food processing industries (Gracha *et al.*, 1993). Previously, Tesfaw *et al.*, (2015) pointed that it contains 91% of water and 10% of dry matter; in addition, its nutritional value can be compared with milk, eggs and meat. Kurtzman, (1967); Hayes Haddad, (1976); Szabova *et al.*, (2013) added that Oyster mushroom is rich in organic components including: vitamins (thiamine, riboflavin, folic and niacin), proteins and minerals, and is considered to be intermediate between that of vegetables and animals. Moreover, it contains all the 9 essential amino acids. This type of mushroom contains high of potassium to sodium ratio, which makes it an ideal food for patients suffering from hypertension and heart diseases (Dehariya *et al.*, 2013). According to Bano *et al.*, (1993); Cohen *et al.*, (2002), cultivation of Oyster mushroom offers the most feasible and economic method for agro-lignocellulosic wastes biodegradation. These biodegrading technologies can also limit air associated with burning agriculture wastes, as well as to decrease environmental pollution due to these unutilized agricultural wastes. The current study aimed to evaluate the optimal nutritional conditions for mycelial growth and production of Oyster mushroom under the conditions of Upper Egypt.

Materials and Methods

Substrate preparation and inoculation

Two agro wastes substrates including (ScB) and (WS) were obtained from Qena governorate farms, Egypt. The RS and WS were dried and then ground into 0.5-2 cm length pellets, and then soaked separately in water over 2 h. In order to determine the suitable substrates for cultivation of mushroom, three substrates including RS, WS, ScB were used, while the fourth substrate formula was composed of their total combination at a proportion of 1:1, following the method of Bano *et al.,* (1982) with slight modifications. Three replicates were used for each substrate formula, they were cooled to room temperature, and then the bags were inoculated with the mushroom spawn.

Mushroom spawn cultivation

The polyethylene bags of the size 30 x 50 cm were filled with sterilized agro-substrates, and the multilayered techniques were adapted for mushroom spawns cultivation (Baysal *et al.*, 2003). About 5 kg of dry substrate was filled in each bag. After inoculation with the spawn, the bags were kept in the greenhouse where the temperature and humidity were maintained at 25^{O}C and 80-90%; respectively, with sufficient light for 20 d. The polyethylene bags were tear-off after the incubation period, and the spawn runs was completed within 18 d. The beds were maintained till the harvest of the third flush, which was completed in 35 d after spawning.

Biological efficiency and moisture content

The total weights of all the fruiting bodies harvested from the three picking were measured as the total yield of mushroom. The biological efficiency (B.E.) (yield of mushroom per kg substrate on dry weight basis) was calculated by the following formula of Chang *et al.,* (1981):

B.E. (%) = (Fresh weight of mushroom/ Dry weight of substrate) × 100

According to Syed *et al.,* (2009), the moisture content (M.C) of the mushroom was also expressed in percentage, and calculated by the following formula:

M.C (%) = (Weight of fresh sample – weight of dry sample/ Weight of fresh sample) × 100

Nutritional analysis

The Protein content, fat, ash and total carbohydrates were determined with the procedure recommended by Wankhede *et al.*, (1976); AOAC. (1995). The crude fibers and calcium were determined according to Ranganna, (1986). Whereas the mineral contents (K, Mg and Zn) were analyzed by the flam and spectrophotometer in reference to Ezeibekwe *et al.*, (2009), after extraction of the elements in 0.1N HCl acidic solutions.

Mushroom incubation and harvest

According to Tesfaw *et al.,* (2015), the inoculated substrates were kept in an incubator at 28ºC and 70-80% relative humidity (RH) under dark conditions. After the substrates surface was entirely covered with the mushroom mycelium, they were moved to an incubation room having temperature of 24ºC, and kept at RH of about 90% or above.

Finally, the stipe length (mm), the cap diameter (cm), and the Fresh weight (g) were measured at the first, second and the third flush, and then the means were also determined at the end of harvest. These accumulated data were used to calculate total yield and the biological efficacy (B.E.).

Statistical analysis

The experiments were arranged in a randomized complete block design with three replicates.

One-way analysis of variance (ANOVA) was conducted with Duncan's multiple range tests to compare the mean significant differences ($p < 0.05$) among the treatments

Results and Discussion

There are differences recorded concerning the cap diameter of *P. cystidiosus* grown on the different substrates formula (Table 1). The cap diameter is the highest (10 cm) on RS and substrate formula Ws+ Rs, conversely the lowest cap diameter (8 cm) is recorded on using ScB. The WS is recorded to be the superior over than other agro-wastes in mushroom production parameters in agreement with Philippoussis, (2003). Moreover, mixing of the agro-wastes at different ratios enhanced the productivity. Figure 1 demonstrates that the lengths of the Oyster mushrooms stipes are changed with the use of the different agro-waste substrates. The stipe length ranged from 32- 43 mm, while the lowest stipe (32 mm) is recorded on using the substrate formula RS+ ScB (Table 1). The values of the stipe lengths of the Oyster mushroom grown on the different substrate formulas are almost the same or lower than those of other experimental substrates (ScB, RS and WS agro-wastes), while the thickness values are the same or higher, according to Dehariya *et al.,* (2013).

The data in Table 2 show that the fresh weight of each picking of the Oyster mushrooms on the lignocllulosic agro-wastes WS- Rs- ScB substrates is lower than that of the other substrate formulas. The total yields of the Oyster mushroom on using the formula of Ws+ RS+ ScB are of good remarkable quantities. The WS presents the maximum number of fruiting bodies and fresh weights, followed by ScB, whereas the lowest number is recorded by RS. These variations are probably attributed to the difference of the bulk density and constituents of the agro-waste substrates. The yields of mushroom are correlated positively with the cellulosic content, but negatively with the lignin and phenoloic contents, in agreement with the findings of Sivaprakasm (1986).

The maximum yield of *P. cystidiosus* is obtained on using WS (970 g/ 5kg straw) with B.E. of 97%, followed by the yield on WS+ ScB (850 g/ 5kg straw) with B.E. of 85%, while the least yield is recorded on using WS+ RS agro-waste (770 g/ kg straw).

The maximum moisture content is recorded on using the ScB (91%), followed by WS (90%). However, there is a slight variation with the other substrates indicating that the moisture content is independent of the substrate. The performance of the three substrates is also evident by their elevated B.E. values on using the WS followed by RS (Table 2). The recorded time required for harvest of the fruiting bodies in WS is always preceded by the RS or RS combination. Cultivation of the *P. cystidiosus* on similar by-products has manifested variable levels of B.E. According to Mane *et al.,* (2007), these variations are mainly related to the spawn rate, fungal species, and to the supplements added to the substrates. Sharma *et al.,* (2013) pointed out that some of the recorded B.E. of the Oyster mushroom on the common substrates such as RS is 85.5%, but on the leguminous plants the B.E. is 103.8%. The total yield of the Oyster mushroom currently recorded is similar the earlier studies of Raina *et al.,* (2009); Patil, (2012).

The protein, fat, carbohydrates, crude fiber and ash contents of the mature fruiting bodies of *P. cystidiosus* cultivated on different lingocellulosic substrates alone or in combination are shown in Figure 2. The mushroom fruiting bodies produced on WS expressed the highest protein content of 23% on a dry weight basis, followed by WS+ RS (22%). The highest fat content of the Oyster mushroom is recorded on using the WS+ RS combination (3.9%), followed by ScB (2.5%). The current protein and fat contents are similar to the earlier studies of Patil and Dakorem, (2007); Patil *et al.,* (2008). The maximum carbohydrate content of the mushroom is 67% in fruiting bodies cultivated on ScB, whereas the least is 51% on cultivation on the WS + ScB, which is consistent with the results of Patil *et al.*, (2008). The highest records of crude fiber is observed on ScB (702%), followed by WS formula (6.9%). The other agro-wastes alone or in combination also yielded appreciable levels of crude fiber. These results are in agreement with the earlier findings of Khyadagi *et al.*, (1998); Singh *et al.*, (2003); Bhattacharjya *et al.*, (2014). The protein content usually ranges between 20- 30% on a dry weight basis. Patil *et al.*, (2008) revealed that the substrates rich in usable nitrogen may be a considerable factor in enhancing the mushroom yield and quality, in addition to the mushroom spices with respect to the bioconversion and bioaccumulation efficiencies.

Table.1 Represent the data of caps diameter, length of stipes and the fresh weights after 3 flushes at the winter season

Agro-waste substrates	Cap diameter (cm)	Fresh weight (g)/ bag dry straw	Length of the stipe (mm)
WS	9.5	410	35
RS	10	355	33
ScB	8	380	34
ScB+ WS+ RS	10	390	36
WS+ RS	10	370	43
RS+ ScB	9	395	32
ScB+ WS	9	400	41

Table.2 Effect of the agro-waste substrates on the total yield, B.E. and moisture content of the Oyster mushroom

Agro-waste substrates	1 picking	2 picking	3 Picking	Total yield (g)/	Bio efficiency (B.E)	Moisture %
(WS)	440	230	200	970	97	90
(RS)	355	275	150	780	78	88
(ScB)	380	260	170	810	81	91
WS+ RS+ ScB	390	240	160	790	79	89
RS+ ScB	395	230	155	780	78	89
ScB+ WS	400	310	140	850	85	88
Ws+ Rs	370	240	160	770	77	89

Fig.1 The oyster mushroom growth bag

Fig.2 Effects of the different agro-wastes on the protein, carbohydrates, fat, fiber and ash contents of the Oyster mushroom

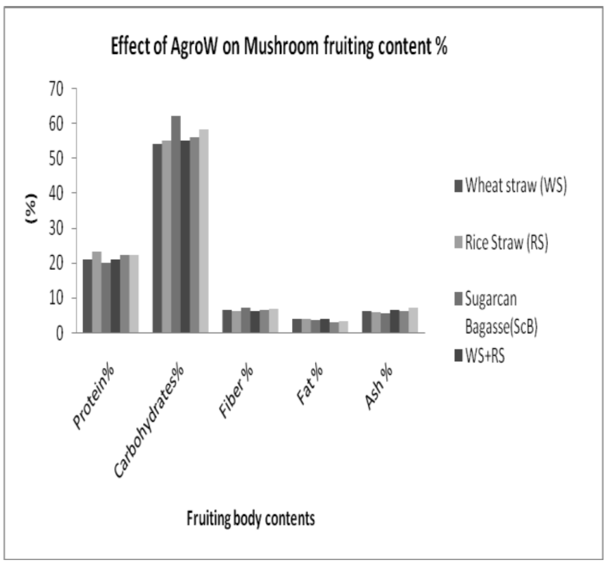
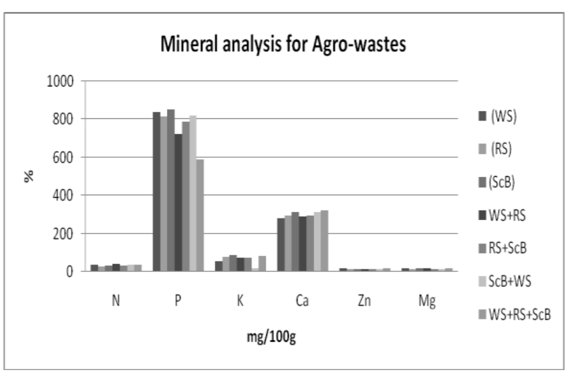


Fig.3 Effects of the agro-wastes substrates on the N, P, K Mg, Zn and Ca contents (mg/ 100g) of the Oyster mushroom



The maximum ash content of Oyster mushroom is found on using the combination of Ws+ Rs+ ScB (7.5%), followed by mushroom grown on WS+ RS (6.5%), similar results were reported by Patil *et al.,* (2001). In a previous study, Quimio, (1987) reported that cellulose rich organic substances are good substrates for the cultivation of mushroom. Agro-waste substrates with high lignin and phenolic contents decrease the activity of cellulose, however less lignin content would enhance the enzyme activity and thus ensure higher yield of the mushroom (Sivaprakasam, 1980). The highest calcium (Ca) content is recorded when Oyster is grown on substrate formula of Ws + Rs (310 mg/ 100g), followed by ScB +WS (280mg/ 100g), as clear in Figure 3. The phosphorus (p) content is maximum on using the ScB (850mg/ 100g), whereas the lowest content is observed in the formula of RS+ WS (722mg/ 100g), similar amount of P was also recorded by Caglarirmak (2007). The highest contents of Zink (Zn) and Magnesium (Mg) in the mycelium of Oyster mushroom is achieved on WS (18mg/ 100g) substrate. The lowest potassium (K) value is observed from the formula of WS+ RS, and is significantly lower than the other substrates formula. In the case of nitrogen (N), the maximum content is achieved on using the substrate formula WS+ RS (39 mg/ 100g). These current results for the K, P and Ca contents are in consistent with the previous studies of Ahmed *et al.,* (2009); Patil *et al.,* (2010); Patil *et al.,* (2012),

According to the data of all agro-wastes estimated in this study, it can be concluded that *P. cystidiosus* in Upper Egypt can grow in a wide range of temperatures, and on several substrates. The best growth is recorded on WS then RS, and then on ScB. Accordingly, growing the Oyster mushroom on WS or ScB is less expensive, due to the availability of these agro-wastes in the surrounding environment. The use of the agro-wastes such as; ScB, RS and WS for Oyster mushroom cultivation is a new way of handling the plantations wastes.

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