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Evaluation of some organic substrates for the growth and yield of oyster mushroom *Pleurotus ostreatus* (Jacq.Fr.) Kumm in southeast Nigeria

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Abstract

Four organic substrates; Mahogany Sawdust (MSD), Corn Cobs (CC), Oil Palm Fruit Fibre (OPFF) and Rice Bran (RB) were evaluated for their effects on growth and yield of *Pleurotus ostreatus* (Jacq.Fr.) Kumm. The completely randomized experimental design was adopted for the study with 4 treatments replicated 10 times. Results on mean number of days for spawn run, primordial formation and formation of fruit body were 19.90 ± 0.28 (CC)- 25.20 ± 0.29 (MSD), 45.10 ± 0.28 (CC)- 47.90 ± 0.23 (MSD) and 56.50 ± 0.22 (CC)- 59.40 ± 0.27 (MSD), respectively. Similarly, height of stipe, diameter of stipe and diameter of pileus ranges were 3.28 ± 0.13 (MSD)- 3.51 ± 0.06 (RB), 1.26 ± 0.06 (OPFF)- 1.39 ± 0.05 (RB) and 4.08 ± 0.05 (CC)- 4.70 ± 0.04 (OPFF), respectively. Fresh weights (g), dry weights (g) and biological efficiency were 10.20 ± 0.31 (OPFF)- 11.05 ± 0.14 (MSD), 3.18 ± 0.15 (CC)- 3.38 ± 0.13 (RB) and 4.14 ± 0.14 (OPFF)- 4.42 ± 0.06 (MSD), respectively. Results on mushroom growth showed that CC took the least duration for full mycelial colonization and the longest duration occurred on MSD. There were significant differences ($P \leq 0.05$) between the durations required for primordia formation among the four organic substrates. The results on mushroom yield showed that mean fresh weights of harvested mushrooms varied from 10.20 ± 0.31 g on OPFF to 11.05 ± 0.14 g on MSD. There were no significant differences ($P \geq 0.05$) in the biological efficiency of mushrooms grown on MSD, CC and OPFF substrates. Considering the growth and yield attributes RB was observed to be the best substrate and could be used for commercial production of *P. ostreatus* among the various organic substrates used in this study.

Key words: *P. ostreatus*, oyster mushroom, yield, organic substrates

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INTRODUCTION

Pleurotus ostreatus (Jacq.) P. Kumm (family Pleurotaceae) has its origin from China but now

of worldwide importance (Wojewoda, 2003). It grows on logs of wood and branches of trees such as, common walnut, birch-tree, hornbeam, willow, poplar, beech and palm trees. It comes

second behind *Agaricus bisporus* as the most economically important cultivated edible mushroom (Sanches, 2010). *Pleurotus ostreatus* (oyster mushroom) has versatile ability to degrade ligno-cellulosic biomass of plant wastes and utilize them for growth and development. This renders such biomass suitable as animal feed and crop manure (Maher, 1991; Ortega *et al.*, 1992). Oyster mushroom contains readily metabolizable vitamins, minerals and protein (Jandaik and Goyal, 1995; Caglarirmak, 2007). Protein from mushroom is intermediary to those from vegetables and animals and is of higher quality due to the availability of all the essential amino acids (Kurtzman, 1976; Purkayastha and Nayak, 1981). *P. ostreatus* is relevant pharmacologically as it has been reported to have hypoglycemic, hypolipidemic and hypocholesterolemic activities (Chorvathora *et al.*, 1993; Anandhi *et al.*, 2013; Jayasuriya *et al.*, 2012). It is also reported to contain antineoplastic (Patel and Goyal, 2012; Siegel *et al.*, 2015), antioxidative (Arbaayah and Kalsom, 2013; Chowdhury *et al.*, 2015; Elbatrawy *et al.*, 2015), antimicrobial (Ahmad *et al.*, 2014; Meza-Menchaca *et al.*, 2015; Younis *et al.*, 2015) and antiviral (Santoyo *et al.*, 2012; Krupodorova *et al.*, 2014) principles. Oyster mushroom is utilized extensively for catalyzation of cumbersome chemical conversions and bio-bleaching in the paper industry, dye de-colorization in textile industry and bio-remediation (Park *et al.*, 2007). Organic substrates of plant origin are available in good quantity in Nigeria, deploying them for mushroom farming ultimately makes them useful, thereby greatly empowering farmers and engendering food security. Therefore, the objective of the present work is to determine the suitability of some organic substrates of plant origin for cultivation of oyster mushroom.

MATERIALS AND METHODS

Source of materials

The substrates namely: Oil palm fruit fibre and corn cobs were sourced locally from Imo State, Nigeria. Rice bran was collected from Awgu town in Enugu State while the Mahogany sawdust was obtained from sawmills in Nsukka, Enugu State. *P. ostreatus* spawn was obtained from the National Biotechnology Development Agency (NABDA) Laboratory, University of Nigeria, Nsukka.

Substrate preparation, spawning and incubation

A modified method of Chiejina and Osibe (2015) was adopted for substrate preparation. The chopped oil palm fruit fibre, mahogany sawdust, ground corn cobs and rice bran were placed on a long flat table after soaking in tap water overnight to reduce the moisture content of the substrates. Two hundred and fifty grams (250 g) of each of the substrates was filled into different heat resistant polyethylene bags measuring 17.5x15 cm. Thereafter, the bags were steam heated to about 80 °C in a pressure pot two third filled with water and the temperature maintained for 2 hours. The bags were drained and allowed to cool at temperature of 27°C ± 3°C. Under aseptic conditions, the bags were randomly picked and spawned with 25 g of the *P. ostreatus* spawn. Thereafter, the mouth of each bag was tied with rubber band and perforated at the sides to allow cross-sectional ventilation after which they were incubated at temperature range of 28-30 °C.

Fructification and harvesting

Spawned bags were transferred to a growing room for mycelia colonization of the substrates. At the emergence of primordial (pin heads), the tops of the bags were partially opened, and spawns were humidified with water using a hand sprinkler (Chiejina and Osibe, 2015). The pin heads subsequently developed into fruit bodies and were harvested 10-15 days later.

Data collection

The following data were collected: chemical composition of substrates (before cultivation), duration for spawn run/mycelia colonization, duration for primordia/ pin heads formation, fruit body yield (stipe height, stipe diameter, pileus diameter, fresh and dry weights of harvested mushroom) and biological efficiency (B.E).

Analytical methods

All chemical analyses were carried out using standard methods (Goering and Soest, 1970; Ilukor and Olukor, 1995; AOAC, 2002). Weights of mushroom were determined using Furi electronic weighing balance. Biological Efficiency (BE: capability of mushrooms to change substrates into fruiting bodies) was determined using the formula adopted by Zenebe *et al.* (2016):

$$B.E = \frac{\text{Fresh weight of harvested mushroom} \times 100}{\text{Dry weight of substrate}}$$

Statistical Analysis

The experimental design used for the study on the four substrates Mahogany sawdust (MSD), Corn cobs (CC), Rice bran (RB) and Oil palm fruit fibre (OPFF) was the completely randomized design (CRD) in ten replications. The data obtained were subjected to a one-way Analysis of Variance (ANOVA) using SPSS version 17.0 and means separated using Least Significant Different (LSD) at 5% level of significance.

RESULTS AND DISCUSSION

The proximate chemical compositions (percentage) of various substrates, MSD, CC,

OPFF and RB used in this work revealed the presence of cellulose, hemicelluloses, lignin, carbon and nitrogen in all the substrates (Table 1). The highest cellulose composition was produced by OPFF (54.88%) while RB (51.36%) contained the least. MSD contained the highest (32.44%) hemicellulose while RB gave the least (28.29%). The lignin content of CC was highest (14.13%) while RB contained the least (11.67%). The carbon content was highest in MSD (48.67%) while that of RB (38.40%) was the least. Meanwhile, RB recorded the highest (1.66%) nitrogen content while MSD produced the least (0.43%). Philippoussis *et al.* (2003) reported carbon (45.65%) in corn cobs, nitrogen (1.33%) in wheat straw, cellulose (47.7%) in oak-wood sawdust, hemicelluloses (37.0%) in corn cobs and lignin (16%) in oak-wood sawdust.

Table 1: Percentage proximate composition of the organic substrates before mushroom cultivation.

Substrates	Cellulose	Hemicellulose	Lignin	Carbon	Nitrogen
OPFF	54.88 ± 0.01 ^a	28.35 ± 0.03 ^c	12.53 ± 0.00 ^b	42.30 ± 0.96 ^b	1.45 ± 0.03 ^a
RB	51.36 ± 0.02 ^c	28.29 ± 0.01 ^c	11.67 ± 0.00 ^c	38.40 ± 0.15 ^c	1.66 ± 0.04 ^a
CC	51.61 ± 0.07 ^b	29.14 ± 0.01 ^b	14.13 ± 0.00 ^a	41.13 ± 0.09 ^{bc}	1.56 ± 0.02 ^a
MSD	53.70 ± 0.07 ^b	32.44 ± 0.01 ^a	12.56 ± 0.01 ^b	48.67 ± 0.22 ^a	0.43 ± 0.00 ^b
LSD	0.72	0.67	0.67	2.90	0.68

Key: Oil Palm Fruit Fibre (OPFF), Rice Bran (RB), Corn Cobs (CC), Mahogany Sawdust (MSD)

The results of the experiment on mushroom growth showed that the mean duration required for spawn run of the organic substrates by the fungal mycelia varied from 19.90 ± 0.28 days to 25.20 ± 0.29 days. The fastest colonization of the wastes 19.90 ± 0.28 days was observed on corn cobs (CC) while the least 25.20 ± 0.29 days was in mahogany sawdust (MSD) (Table 2). This agrees with the findings of Shah *et al.* (2004) who reported that it took 21 days for the mycelia of *P. ostreatus* to fully colonize wheat straw, leaves and saw dust substrates. Mycelial growth is a preliminary step that creates suitable internal conditions for fruiting. Thus, outstanding growth of mycelium is a vital factor in mushroom cultivation (Pokhrel *et al.* 2009). Sharma *et al.* (2013) also reported in their findings that it took 22.40 – 26.00 days for mycelia of *P. ostreatus* to fully colonize rice straw, rice straw + wheat straw, rice straw + paper, sugarcane bagasse and sawdust substrates. The effects of the substrates on duration (days) to primordia formation ranged

from 45.10 ± 0.28 to 47.90 ± 0.23 days for corn cobs and mahogany sawdust, respectively. There was significant difference ($P \leq 0.05$) in the time required for primordia formation among evaluated substrates. The effects of the substrates on fruit body formation showed that the fastest fruit body formation 56.50 ± 0.22 was observed on the rice bran (RB) wastes and the slowest 59.40 ± 0.27 was on the mahogany sawdust (MSD). There was significant difference ($P < 0.05$) on the duration required for fruit body formation grown on the organic wastes (Table 2).

The fastest mycelia colonization 19.90 ± 0.28 days and primordial formation 45.10 ± 0.28 of the *P. ostreatus* in the substrates, was observed on corn cobs (CC) while the slowest mycelium colonization 25.20 ± 0.29 days and primordial formation 47.90 ± 0.23 was seen in mahogany sawdust (MSD). This agrees with the findings of Buah *et al.* (2010) who reported fastest mycelia colonization in corn cobs in this study.

Table 2: Effect of the different substrates on spawn run, primordia and fruit body formation by *P. ostreatus*.

Substrates	Complete spawnrun (days)	Primordia formation (days)	Fruit body formation (days)
OPFF	22.70 ± 0.21b	47.30 ± 0.37ab	57.90 ± 0.28ab
RB	20.10 ± 0.28c	46.10 ± 0.57ab	56.50 ± 0.22b
CC	19.90 ± 0.28c	45.10 ± 0.28b	56.90 ± 0.31b
MSD	25.20 ± 0.29a	47.90 ± 0.23a	59.40 ± 0.27a
LSD (0.05)	1.82	2.54	1.86

Key: Oil Palm Fruit Fibre (OPFF), Rice Bran (RB), Corn Cobs (CC), Mahogany Sawdust (MSD).

The fast mycelia colonization and primordia formation in corn cob substrates could be as a result of the chemical composition of the substrate, particularly because of its high nitrogen content, particle size and or the pore size of the substrate. Chiejina and Osibe (2015) observed that substrates with larger pore size and surface area gave faster mycelium growth rate. The presence of substantial amounts of cellulose and hemicelluloses in various substrates has been reported to support vigorous proliferation of mushroom mycelia. Chiejina and Osibe (2015) reported that growth of mushroom mycelia depends on the nitrogen content of the substrate. Naraian *et al.* (2009) reported that mycelia growth and primordia development depends heavily on the lignocellulosic materials especially the C:N of the substrates. The appreciable amount of nitrogen (1.56 %) in chemical composition of corn cobs in this experiment, explains the very fast substrate colonization and primordia formation. Early fructification of the *P. ostreatus* was observed in the rice bran substrate. The nitrogen content of substrates has been shown to influence quick fructification and productivity of mushrooms (Philippoussis *et al.*, 2007). The early fructification could be as a result of the appreciable amount of nitrogen (1.66 %) in rice bran when compared to the other organic wastes. The results showed that mahogany sawdust (MSD) took the longest time for mycelium colonization. This confirms the findings of Buah *et al.* (2010) who reported longest mycelia colonization of *P. ostreatus* in sawdust substrate. Akinmusire *et al.* (2011) also reported the longest colonization time for *P. pulmonarius* on sawdust. However, contrary to our results, Islam *et al.* (2009) reported faster mycelium colonization and

fructification for *P. ostreatus* on sawdust substrate.

The slow growth of the mushroom on mahogany sawdust could probably be due to the fact that the wood contains particles that inhibit growth of fungi as reported by Davis and Aegeter (2000). Mycelium extension rate has been reported to be related to bioavailability of nitrogen in the cultivation substrate (Philippoussis *et al.*, 2007). The low nitrogen content (0.43%) of mahogany sawdust may have also contributed to the slow growth and fructification of the mushroom in organic substrates. Several reports also show differences in the rates of delignification of substrates by mushroom mycelia which result in variations in the number of days to first fructification (Chitamba *et al.*, 2012).

The results on mushroom yield showed that rice bran produced mushrooms with the highest mean stipe height (3.51 ± 0.06 cm) while mahogany sawdust recorded the least (3.28 ± 0.13 cm) (Table 3). There was no significant difference ($P \geq 0.05$) in the stipe height produced on the various organic wastes. Mushrooms with the largest stipe diameter (1.39 ± 0.05 cm) were observed in rice bran while the least stipe diameter (1.26 ± 0.06 cm) was produced from oil palm fruit fibre. There was no significant difference ($P \geq 0.05$) in the stipe diameter of mushrooms produced from the various substrates evaluated. Oil palm fruit fibre recorded the highest mean pileus diameter (4.70 ± 0.04 cm) while corn cob produced the least (4.19 ± 0.05 cm). Analysis of variance indicated that there were significant differences ($p \leq 0.05$) in the mean pileus diameter of the mushrooms. This result corroborates the findings of Chiejina and

Table 3: Effect of the different substrates on stipe height, stipe diameter and pileus diameter of the harvested mushrooms

Substrates	Height of stipe (cm)	Diameter of stipe (cm)	Diameter of pileus (cm)
OPFF	3.43 ± 0.12	1.26 ± 0.06	4.70 ± 0.04 ^a
RB	3.51 ± 0.06	1.39 ± 0.05	4.40 ± 0.07 ^b
CC	3.35 ± 0.13	1.34 ± 0.07	4.08 ± 0.05 ^b
MSD	3.28 ± 0.13	1.29 ± 0.09	4.21 ± 0.11 ^{ab}
LSD	NS	NS	0.69

Key: Oil Palm Fruit Fibre (OPFF), Rice Bran (RB), Corn Cobs (CC), Mahogany Sawdust (MSD)

Table 4: Effect of the different substrates on fresh weight, dry weight and biological efficiency of the harvested mushrooms

Substrates	Fresh weight of harvested mushroom/bag (g)	Dry weight of harvested mushrooms per bag (g)	Biological efficiency (%)
OPFF	10.20 ± 0.31	3.23 ± 0.12	4.14 ± 0.14
RB	10.53 ± 0.28	3.38 ± 0.13	4.21 ± 0.11
CC	10.37 ± 0.23	3.18 ± 0.15	4.15 ± 0.09
MSD	11.05 ± 0.14	3.34 ± 0.13	4.42 ± 0.06
LSD	NS	NS	NS

Key: Oil Palm Fruit Fibre (OPFF), Rice Bran (RB), Corn Cobs (CC), Mahogany Sawdust (MSD).

Osibe (2015) who reported that oil palm fruit fibre produced the highest pileus diameter for *Lentinus squarrosulus* mushroom. Earlier reports showed that plant oils are stimulatory to mushroom mycelia growth and sporophore production since they are required for cell membrane expansion and consequently results in higher yields (Chiejina and Osibe, 2015). Sharma *et al.*, (2013) reported that oyster mushrooms usually have a cap diameter of 5 to 25 cm at maturity and this agrees with results of the present work. Regarding the mean fresh weight, mahogany sawdust recorded the highest mean fresh weight (11.05 ± 0.14 g) while oil palm fruit fibre produced the least (10.20 ± 0.31 g). The highest mean dry weight (3.38 ± 0.13 g) of *P. ostreatus* was produced by rice bran while the least (3.18 ± 0.15 g) was obtained from those grown on corn cobs. However, there were no significant differences ($P \geq 0.05$) in both the mean fresh weight and dry weights of mushroom grown on the different substrates (Table 4). Weight of mushrooms

irrespective of the nature (fresh or dry) is a vital agronomic parameter for evaluating the potency of fungi as biological agents in conversion of inedible organic wastes directly into palatable human food (Mwita *et al.*, 2011). In this study, mahogany sawdust (MSD) and rice bran (RB) produced mushrooms with the highest mean fresh and dry weights, respectively. This compares with the work of Joshua and Angina (2002), who reported a high yield of *P. ostreatus* on sawdust of *Khaya ivorensis* and *Mansonia altissima*. In a related work, Adjapong *et al.* (2015) reported highest fresh weight of *P. ostreatus* mushrooms grown on sawdust substrate. The high fresh weight observed in mushrooms grown could be due to good aeration and composition of substrates as reported by Adjapong *et al.* (2015). The mean highest dry weight observed in rice bran corroborates the work of Mamiro and Mamiro (2011) who reported highest weight of *P. ostreatus* mushroom grown on sunflower and cotton seed cake supplemented with rice straw

Similarly, Frimpong-Manso *et al.* (2011) reported highest weight of *P. ostreatus* on rice husk and attributed it to physical nature, high porosity and high level of aeration of the rice husk. Cellulose, hemicellulose, lignin and nitrogen contents of substrates as well as the necessary enzyme production by mushrooms are important criteria for yield determination in mushrooms (Philippoussis *et al.*, 2003). The lowest fresh and dry weights recorded for fruit bodies harvested from oil palm fruit fibre (OPFF) and corn cob (CC) could be due to the complex nature of the waste and/or the presence of little or no vital nutrients needed for the mushroom growth in the substrates. The reduction in fresh weight of mushrooms has been associated with the absence of certain specific nutrients in the substrate required by the mushroom for its growth (Tinoco *et al.*, 2001). Table 4 also showed that mahogany sawdust produced mushrooms with the highest mean percentage biological efficiency (4.42 %) while OPFF produced mushrooms with the least (4.14 %). There were no significant differences ($P \geq 0.05$) in the mean percentage biological efficiency of mushrooms grown on the various wastes. Pathania *et al.* (2017) reported biological efficiency of *P. ostreatus* grown on mixtures of apple pomace and wheat straw ranging from 0.091% to 8.80%. The potency of residues bioconversion process and the productivity of the mushroom crop are assessed by the biological efficiency (BE) (Philippoussis and Diamantopoulou, 2011). The observed variable range of BE in this work, may be due to the differences in composition and rate of degradation of the lignocellulosic materials in the wastes by *P. ostreatus* enzymes. Variable range of BE has been reported when different lignocellulosic materials were used as substrates for the cultivation of mushrooms (Liang *et al.*, 2009).

Conclusion

In conclusion, RB produced the best result in terms of growth and yield attributes while MSD was the best in biological efficiency (BE).

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Declaration of conflict of interest:

Authors have declared no conflict of interest.

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