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Effect of Honey, Palm Oil and NPK Fertilizer on the Growth of Oyster Mushrooms Cultivated on Sawdust

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ABSTRACT: The study was carried out to determine the effect of honey, palm oil and NPK fertilizer on the growth of *Pleurotus pulmonarius* (Fr.) Quel. and *Pleurotus ostreatus* (Jack.) Kumm. on *Celtis zenkeri* Engl. sawdust. Composted sawdust was supplemented with honey, palm oil and NPK fertilizer at 0%, 2%, 4%, 6% and 8% on the dry weight basis and inoculated with 5% spawn. The substrate amended with 4% palm oil was fully colonized by *Pleurotus ostreatus* and *P. pulmonarius* after 26.0±2.1 and 32.2±1.3 days, respectively. The mushroom took shorter time (43 days) for primordial emergence at 6% palm oil. Average of 55 fruitbodies of *P. ostreatus* was harvested from 4% palm oil supplemented sawdust. The best mushroom yield was recorded from 8% palm oil (278.5 ±5.0 g) and 8% honey (270.1±2.7 g). The highest biological efficiency was observed at 8% palm oil (77.36±1.5%) and 8% honey (75.03±2.5%) for *P. ostreatus*. The two mushrooms species performed significantly (p=0.05) with palm oil and honey supplementation. The best performance was observed at 6% and 8% supplementation with palm oil and honey. The research has shown that supplementation of sawdust substrate with honey and palm oil at 4 - 8% can be a good formulation for commercial cultivation of oyster mushrooms.

Keywords: *Pleurotus pulmonarius*, *Pleurotus ostreatus*, Supplemented substrate, Mushroom

Introduction

Mushroom cultivation is a worldwide practice that utilizes almost all agricultural and agro-industrial residues as substrates (Chang, 1999). Mushroom production in Nigeria and the world at large is a growing commercial venture. Mushrooms have been considered as one of the most functional food with many well-known therapeutic applications (Deepalakshmi and Mirunalini, 2014).

In Nigeria, many mushrooms are highly prized, not only as food but also in traditional medicine (Alabi, 1990). Oyster mushrooms (*Pleurotus* spp.) are edible mushrooms that are commercially cultivated worldwide. *Pleurotus* species are the easiest and least expensive commercial mushroom to grow because they are well known for the conversion of crop residues to food protein (Banik and Nandi, 2004). They have the ability to grow over wide range of temperatures, utilizing various lignocellulosic (Sanchez, 2010). Oyster mushrooms have high nutritional value; they are important source of quality protein, carbohydrates, vitamins, calcium and iron. The nutritional advantages of mushrooms include a low content of calories and a high content of proteins, minerals and dietary fiber (Beluhan and Ranogajec, 2011). Oyster mushrooms are rich in Vitamin C, B complex and mineral salts required by the human body (Randive, 2012). Sawdust is a major waste product from timber industry in Nigeria, and is readily exploitable for oyster mushroom cultivation (Ayodele and Akpaja, 2007). Sawdust is poor in nitrogen and phosphorus, and as a result various attempts have been made to grow *Pleurotus* species on supplemented sawdust wastes (Onyango *et al.* 2011). Supplementation of substrates with different levels of carbonates and nitrogen-based additives has been shown to enhance mushroom production (Isikhuemhen *et al.*, 1999; Royle 1996). Different plant species sawdusts, even under supplementation, vary in performance when used for mushroom cultivation. Sawdust from *C. zenkeri*, is common in Nigeria; to the best

of our knowledge, it has never been studied for oyster mushrooms cultivation in Nigeria. This study was tested on sawdust of *C. zenkeri* supplemented with honey, palm oil or NPK fertilizer for the mushrooms, *Pleurotus ostreatus* and *P. pulmonarius*.

Materials and methods

The study was conducted at the African Centre for Mushroom Research and Technology Innovations (ACMRTI), University of Benin. Pure cultures of *P. ostraetus* and *P. pulmonarius* were obtained from ACMRTI. The spawn of the mushrooms were prepared from the pure culture using sorghum grains in 250 ml bottles. Sawdust of *C. zenkeri* obtained from a sawmill factory near Uselu market in Benin City was thoroughly mixed with water and 20% wheat bran. The mixture was composted for 14 days with turning every two day (Quimio *et al.*, 1990). The composted substrate was mixed with 1% CaCO₃ and 1% CaSO₄ on oven dry weight basis. The moisture content of the composted substrate was adjusted to 70% with addition of sterile water. 1.2 Kg sawdust of the substrate supplemented with honey, palm oil and NPK fertilizer at 0%, 2%, 4% 6% and 8% respectively were packed into polythene bags.

The un-supplemented (0%) substrates were used as control. The prepared substrates were loaded into polythene bags measuring 15 x 30 cm each. Treatments were replicated three times. The bags were necked with PVC pipe measuring 5 cm x 3 cm, plugged with cotton wool, covered with foil paper and pasteurized. The pasteurized bags were inoculated with spawn of the mushrooms at the rate of 5%, and incubated at room temperature in the dark. The mycelia growth of the mushroom was observed until complete colonization. Colonized bags were then opened for sporophore formation.

Statistical analysis: The data obtained for different parameters were statistically analysed to determine significant differences in yield among the treatments. All the data collected on different parameters were statistically analysed by using computer software SPSS 16.0. The mean values of all the parameters were evaluated and analysis of variance was performed by the ANOVA. The significance of the difference among the treatment means was estimated by the least significant difference (LSD) test at 5% level of probability.

Results

The results revealed that the time required for the complete mycelium colonization of the test substrates varied significantly for the different treatments tested (Table 1). The longest time required to complete mycelium running was observed in 0 % palm oil supplemented for *P. pulmonarius* (48.20 ±1.0) and *P. ostraetus* (39.10 ±1.4) days respectively. In both strains, the shortest time (26.0±2.1 days) for complete colonization was recorded in 4% palm oil supplemented sawdust. NPK treatment performed poorly as the two species took much time (48.20 ±1.0 days) at 0 % and (50.4 ±4.2 days) at 2 % for *P. pulmonarius* to complete their mycelia colonization.

Table 1: Time (days) for complete mycelia colonization of supplemented sawdust by *P. pulmonarius* and *P. ostraetus*

% Level of supplementation	<i>Pleurotus pulmonarius</i>			<i>Pleurotus ostraetus</i>		
	Palm oil	NPK fertilizer	Honey	Palm oil	NPK fertilizer	Honey
0	*39.10±1.4 ^a	48.2±1.0 ^a	41.64±2.4 ^a	39.10±1.4 ^a	34.44±2.7 ^a	37.40±3.1 ^a
2	34.10±2.1 ^b	50.4±1.2 ^a	38.22±0.0 ^a	28.40±1.1 ^b	32.10±0.2 ^b	30.00±1.2 ^b
4	32.20±1.3 ^b	52.11±2.1 ^a	37.7±1.4 ^a	26.3±0.3 ^b	36.32±2.3 ^a	34.31±1.7 ^a
6	34.5±2.3 ^{ab}	ND	38.0±1.3 ^a	28.22±1.7 ^b	ND	28.0±2.3 ^b
8	36.41±3.1 ^a	ND	36.1±0.3 ^b	30.24±2.0 ^b	ND	29.41±2.0 ^b

*Mean ± SD value of three replicates; values with same superscript are not significantly different at P≤0.05. ND: Not detected.

Table 2 shows that there was variation in terms of sporophore formation for the different treatments. The highest time for sporophore formation was recorded for 2 % honey (61.3 ±3.2 days) followed by 6 % honey (58.0 ±3.0 days) and least for 2 % honey (37.20 ±0.2 days). 6 %, 8 % palm oil and 8 % honey significantly enhanced the

yield of the mushrooms under study. NPK at levels higher than 2 % inhibited the growth and yield of the mushrooms.

Table 2: Time (day) for sporophore formation of supplemented sawdust by *P. pulmonarius* and *P. ostreatus*

% Level of supplementation	<i>Pleurotus pulmonarius</i>			<i>Pleurotus ostreatus</i>		
	Palm oil	NPK fertilizer	Honey	Palm oil	NPK fertilizer	Honey
0	*50.2±1.2 ^a	50.0±1.2 ^a	50.2±1.2 ^b	48.2±2.1 ^a	48.2±2.1	48.2±2.1 ^{ab}
2	42.0±2.0 ^b	51.1±3.1 ^a	61.3±3.2 ^a	41.0±0.2 ^b	ND	37.2±0.2 ^c
4	45.0±0.1 ^b	52.0±2.2 ^a	57.2±2.7 ^a	46.8±2.1 ^a	ND	45.22±2.4 ^b
6	44.0±2.9 ^b	ND	58.0±3.0 ^a	42.0±3.0 ^b	ND	50.0±2.0 ^a
8	48.0±3.1 ^a	ND	49.0±3.9 ^b	44.3±0.3 ^b	ND	51.0±0.3 ^a

*Mean ± SD value of three replicates; values with same superscript are not significantly different at P≤0.05. ND: Not detected.

The highest mushroom yield was recorded for *P. ostreatus* at 8 % palm oil (278.50 ±5.0 g) followed by 8 % honey (270.10 ±2.7 g). The least yield (228.10 ±1.0 g) was recorded in the non-supplemented sawdust (Table 3). Plates 1 & 2 show the effects of the supplements on the sporophore yields of *P. pulmonarius* and *P. ostreatus* respectively.

Table 3: Mushroom yield (fresh and dry weight in gram) from supplemented sawdust

% Level of supplementation	<i>Pleurotus pulmonarius</i>			<i>Pleurotus ostreatus</i>		
	Palm oil	NPK fertilizer	Honey	Palm oil	NPK fertilizer	Honey
0	*228.0±0.1 ^d (41.53±1.0)	228.0±0.1 (41.5±10)	228.0±0.1 ^b (41.53±1.0)	228.0±0.1 ^c (41.53±1.0)	ND	228.0±0.1 ^c (41.53±1.0)
2	242.0±2.0 ^b (58.78±2.0)	ND	224.3±2.3 ^c (36.00±1.1)	238.0±2.0 ^d (6.2±1.6)	ND	222.1±0.3 ^c (32.66±1.1)
4	234.2±2.4 ^c (57.5±0.6)	ND	225.2±0.7 ^c (37.53±0.0)	260.24±0.4 ^b (18.8±1.4)	ND	222.3±0.7 ^c (30.90±0.0)
6	261.70±5.0 ^a (59.38±0.9)	ND	227.0±2.1 ^b (40.36±1.2)	249.8±1.7 ^c (60.19±0.2)	ND	227.0±2.1 ^b (40.35±1.2)
8	237.2±2.4 ^c (58.02±0.8)	ND	238.3±2.9 ^a (57.2±0.9)	278.5±5.0 ^a (90.82±2.6)	ND	270.1±2.7 ^a (73.59±1.9)

*Mean ± SD value of three replicates; value in parenthesis is dry weight. Values with same superscript are not significantly different at P≤0.05. ND: Not detected.



Plate 1: Fruit bodies of *Pleurotus pulmonarius* at 6% palm oil and 8% honey.



Plate 2: *Pleurotus ostreatus* at 8% palm oil and 8% honey.

The highest biological efficiency (77.36 ± 1.52 and 75.03 ± 2.51) was recorded from 8% palm oil and 8% honey respectively, for *P. ostreatus*. Although the yield and biological efficiency for honey and palm oil at 2% were lower, they were however not significantly different from treatment at 4% for the mushrooms tested (Table 4).

Table 4: Biological efficiency (%) of Oyster mushroom cultivated on supplemented sawdust

%	<i>Pleurotus pulmonarius</i>			<i>Pleurotus ostreatus</i>		
	Palm oil	NPK fertilizer	Honey	Palm oil	NPK fertilizer	Honey
0	63.33 ± 1.2^d	63.33 ± 1.2^a	63.33 ± 1.2^a	61.69 ± 2.1^b	62.27 ± 2.1	62.27 ± 2.1^b
2	67.2 ± 2.1^b	ND	62.31 ± 5.8^a	66.10 ± 3.0^b	ND	61.69 ± 4.3^b
4	65.1 ± 0.85^b	ND	62.56 ± 2.6^a	72.27 ± 2.1^a	ND	61.75 ± 1.5^b
6	72.69 ± 2.1^a	ND	63.06 ± 3.0^a	68.56 ± 3.2^a	ND	63.05 ± 2.6^b
8	65.89 ± 2.3^b	ND	66.19 ± 3.1^a	77.36 ± 1.5^a	ND	75.03 ± 2.5^a

*Mean \pm SD value of three replicates; values with same superscript are not significantly different at $P \leq 0.05$. ND: Not detected.

Discussion

This study reveals a reduction in the time of mycelia colonization as the concentration of NPK increases. This trend is in line with earlier report of Shalahuddin *et al.*, 2018 that the average time for mycelia colonization and primordial formation for oyster mushrooms varied significantly with NPK treatment. Ayodele and Okhuoya (2013) reported that NPK at 0.05% level gave optimal supply of nitrogen, phosphorus and potassium as higher levels became inhibitory to mycelial growth and mushroom yield. The poor growth and yield recorded for NPK supplementation may be due to the inability to use inorganic form of N, P and K at levels above 2% in the substrate. The time required for the sporophore formation, which was between 37-61 days from spawning was comparable with other similar studies (Iwuagwu *et al.*, 2020; Islam *et al.*, 2009) where sporophores appeared around 18-30 days and 28-34 days on different substrates. Variation in mycelia growth rate, colonization and primordial initiation has also been observed when other *Pleurotus* mushroom species were grown on a wide range of substrates (Islam *et al.*, 2009; Hwang *et al.*, 2015).

Supplementation with palm oil gave the best time range for primordial emergence for *P. pulmonarius* (42-44 days) and *P. ostreatus* (41-42 days) respectively. Ayodele and Akpaja (2007) reported that supplementation of sawdust with 20% oil palm fibres enhanced the mycelia and sporophore yield of *Lentinus squarrosulus*. Nutritional composition of substrate is a crucial factor in determining how mycelia growth and primordial initiation occur (Stamets, 2005). Narain *et al.* (2008) and Kimenju *et al.* reported that mycelia growth and primordial development is dependent on the lignocellulosic materials especially the carbon: nitrogen ratio.

The 6%, 8% palm oil and 8% honey significantly at $p=0.05$, enhanced the yield of the mushrooms under study. NPK at levels higher than 2% inhibited mycelia colonization of substrate and fruit body yield. Ayodele and Okhuoya (2013) reported that application of NPK fertilizer at rate higher than 2% lowered fruit body formation in *Psathyrella atroumbonata*. Supplements or additives usually change the decomposition rate and also the sequence of decomposition of substrate components (Zadrazil, 1993).

Liang *et al.* (2009) variable ranges of biological efficiency (BE) have been reported when different lignocellulosic materials were used as substrates for cultivation of oyster. Girmay *et al.* (2016) reported BE of 74.17% was obtained from cotton seed waste, 9.73% from sawdust, 34.22% from paper waste and 35.88% from wheat straw. The result obtained are also in line with the report of Islam and Raiz (2017) which recorded BE of 92.9% (cotton waste), 83.34% (*Chenopodium album*), 75.77% (*Chenopodium album* and cotton waste), and

71.48% (*Amaranthus viridis*). The results for BE were however lower than the report of Biswas and Biswas, (2015) who observed BE of 104%, 101% and 102% from substrates supplemented with 5% rice bran, 5% wheat bran and 5% maize flour respectively. Again, Ghosh and Biswas, (2012) reported that supplemented substrates with palm oil gave BE of 94% at 0.01 mg/ml and 81.2% at 0.02 mg/ml. The lipid in palm oil must have been efficiently utilized by the mushrooms to achieve the high yield and BE values recorded.

Oyster mushrooms can be economically cultivated on sawdust supplemented with palm oil and honey. The report has shown that supplementation of sawdust substrate with honey and palm oil can be used to increase and influence mycelia growth, mushroom yield and biological efficiency of oyster mushrooms. The study is a part of the effort to utilize abundant agro-industrial wastes for the production of edible and medicinal mushrooms in Nigeria. Further work is therefore recommended at developing a better sawdust substrate supplementation that will produce improved and economic yield of oyster mushrooms.

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