Nutritional values of oyster mushroom (*Pleurotus ostreatus*) (Jacq. Fr.) Kumm. cultivated on different agricultural wastes

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Abstract: In this study, different agricultural wastes such as (*Oryza sativa* straw, *Gossypium hirsutum* wastes and *Milicia excelsa* sawdust were used for the cultivation of oyster mushroom *Pleurotus ostreatus* (Jacq. Fr.) Kumm. Different agricultural wastes were mixed evenly with *Oryza sativa* bran additive in other to enhance mycelial growth of this fungus. High moisture content (93.43%), crude protein (28.02%), fat contents (8.72%) and fiber contents (17.42%) of *P. ostreatus* were all recorded with cotton waste substrate supplemented with 20% rice bran. Results showed that this mushroom was deficient in vitamin A, but contained appreciable amount of vitamin B1, B2, B3, B6 and D. The average highest mushroom yield (6.42g) was recorded on cotton waste substrate +20% rice bran and the lowest mushroom weight (3.62g) was recorded on rice straw without rice bran. Highest biological Efficiency (B.E) was 93.6% while the lowest was 74.0%. Cotton waste was found to be the best substrate that supported the growth of this fungus.
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1. Introduction

Pleurotus species otherwise known as oyster mushrooms are fungi with distinct fleshy fruiting bodies. The genera of pleurotus are delicacies in different parts of the world because of their excellent flavor and taste (Shah *et al*., 2004; Chang and Miles, 1992; Jonathan and Esho, 2010).

Among the genera of *Pleurotus, P. ostreatus* is the most popular and widely cultivated in different regions of the world. Mushroom farmers usually make million of dollar from this single species(Jonathan *et al*,2008).All oyster mushrooms belong to the class Basidiomycetes, subclass Hollobasidiomycetidae, order Agricales and family pleurotaceae (Alexopolous *et al*, 1996). Like any other mushrooms,they could be found in the wild or cultivated artificially by mushroom farms. *P ostreatus* is cultivated in both tropical , subtropical and temperate regions of the world. This mushroom has ability to grow on varieties of agro-industrial wastes (Shal *et al*, 2004; Jwanny *et al*., 1995; Jonathan and Adeoyo, 2011). All Pleurotus species are consumed for their nutritive as well as medicinal values (Agrahar-Murugkar & Subbulakshmi, 2005; Jonathan and Esho, 2010; Jonathan *et al*., 2011). *P. ostreatus* is appreciated more than other Pleurotus species because of its delicious taste; and high quantities of proteins, carbohydrates, minerals (calcium, phosphorus, iron) and vitamins (thiamin, riboflavin and niacin) as well as low fat (Manzi *et al*., 1999; Kurtzman, 2005). *Pleurotus* species also play important role in biodegradation of wastes (Jonathan *et al*, 2012a).

Fasidi *et al*(2008) reported that *P.ostreatus* could be cultivated on a large number of lignocellulose wastes having cellulose, hemicellulose and lignin. These agro-wastes include cotton waste, sugar cane bagasse, wheat straw, rice straw, paper waste, sawdust of different tree species, stalk and leaves of maize and millet, corncobs, pea nut shells, dried grasses and many other wastes. It can also be cultivated by using agro industrial wastes like paper mill sludges, coffee byproducts, tobacco waste, apple pomace etc (Yildiz, 2003; Fasidi *et al*., 2008; Jonathan and Esho, 2010). *P. ostreatus* is common in Europe and North America, and can now be commonly found in their supermarkets (Hawksworth *et al.*, 1995). A single sporocarp is a large structure with a grey to grey-brown cap that grows to around 5-15cm in its longest dimension. The stalk of *P. ostreatus* is generally white, short and eccentric. Generally, this fungus is fragile, relative to the button mushroom (*A.bisporus*). The normal fruiting period is during the autumn and winter months when the temperature is approximately 15-20C (Leigh, 1994). *Pleurotus* spp. accounted for 14.2 % of the total world production of edible mushroom in 1997 (Chang and Miles, 1992). Mushrooms generally had abilities of producing exo-polysaccharides and other secondary metabolites (Aina *et al*,2012; Jonathan and Adeoyo, 2011). Also, they played important roles in the management of organic wastes whose disposal has become a problem(Jonathan and *et al*,2012a).A threat to *Pleurotus spp* fruitbody production is *Trichoderma viride* the causative agent of green mould. Control measures targeted at the invasion of *Trichoderma sp* in the growth substrates used for edible mushrooms were suggested by (Jonathan *et al*, 2012b). Gbolagade *et al* (2006a), reported that the growth of mushrooms could be affected by relative humidity of the atmosphere, temperature, pH and light intensity when it was grown on lignocelluloses wastes. Therefore, this study is aimed at determining the yield, the chemical compositions, the mineral element contents of *Pleurotus ostreatus* cultivated on selected agro-industrial wastes..

2. Materials and Method

The agro-wastes namely *Gossypium hirsutum* waste, *Oryza sativa* straw and *Oryza sativa* bran were collected from International Institutes of Tropical Agriculture (IITA) while *Milicia excelsa* sawdust were collected from Bodija plank market Ibadan. The fungus was collected from the stock culture in Mycology section Department of Botany and Microbiology, University of Ibadan. Pure culture of the fungus was maintained on potato dextrose agar (oxoid) plates.

2.1.Substrate preparation

Gossypium hirsutum waste, *Oryza sativa* straw and *Milicia excelsa* sawdust were soaked separately in water for moisture absorption. The substrates were mixed evenly with rice bran supplement at different concentration (20%, 15%, 10%, 5% and 0% respectively. They were placed in transparent jam bottles (250ml) in replicate of 3. The opening of the bottles were covered with aluminum foil and sterilized in an autoclave at 121^{0}C for 30minutes and allowed to cool at room temperature. After sterilization, each of the experimental bottles was inoculated at the centre of the substrate with 10g of pure culture of *P. ostreatus* under aseptic condition and the bottles recapped with aluminum foil. They were kept in the spawn room, in a dark at 25 - 30^{0}C and 90% Relative Humidity. The bottles were inspected regularly to ensure proper fructification of mushrooms and to avoid contamination of substrates by unwanted microorganisms. The spawn was allowed to run for 21 days. Substrates were completely colonised by the mushroom mycelia. When the mycelia had fully covered the substrates bottles (spawn completed), bottles were opened and regularly watered for fructification. They were harvested after after 27 days of spawning. The weights of the harvested *P. ostreatus* were taken and recorded. The pileus and stipe of the mushrooms were measured in cm using metre rule. Fruit bodies of the mushroom were harvested in flushes.

2.2 Yield and Biological Efficiency

Weights of all the fruiting bodies harvested from bottles were recorded as total yield of mushroom. The biological efficiency B.E was calculated using the formula given by Chang *et al.*, (1999).

B.E was determined as the ratio of fresh mushrooms harvested (g) per dry substrate (g) and expressed as a percentage as shown by the formula.

$$\text{Biological efficiency BE} = \frac{\text{Weight of fresh mushroom harvested (g}}{\text{Weight of dry substrate (g)}} \times \frac{100}{1}$$

2.3 Proximate Analyses

Analysis of moisture, protein, fat, crude fiber, total carbohydrates, ash and vitamins contents were done by standard methods (AOAC, 1995) for *P. ostreatus* fruiting bodies.

2.4 Statistical Analysis

All the data obtained from this study were analyzed using the analysis of variance (ANOVA) procedure by Statistical Analysis System (SAS) and means separation was by Duncan's Multiple Range Test (DMRT) at p = 5%.

Table 1. Measurement of morphological characters of *Postreatus* grown on different agro-wastes

Substrates	***Additive (%)***	***Length of pileus (cm)***	***Length of stipe (cm)***	***Diameter of pileus (cm)***	***Diameter of stipe (cm)***	***Width of stipe (cm)***
Cotton waste	Control	6.2±0.087	3.1±0.133	3.1±0.058	1.6±0.067	3.2±0.058
	5%	6.7±0.145	3.7±0.173	3.3±0.145	1.9±0.087	3.3±0.072
	10%	7.1±0.177	4.0±0.058	3.6±0.088	2.0±0.033	3.7±0.058
	15%	7.3±0.177	4.2±0.145	3.7±0.088	2.3±0.073	4.0±0.058
	20%	8.0±0.231	5.1±0.349	4.0±0.116	2.2±0.187	4.3±0.088
Rice straw	Control	5.3±0.208	2.9±0.088	2.6±0.030	1.5±0.044	3.6±0.153
	5%	5.7±0.186	3.0±0.088	2.9±0.092	1.5±0.044	3.2±0.100
	10%	6.5±0.088	3.4±0.219	3.3±0.038	1.6±0.030	3.5±0.145
	15%	7.2±0.088	4.3±0.153	3.6±0.044	2.1±0.030	3.9±0.058
	20%	7.9±0.120	5.1±0.058	3.9±0.060	2.4±0.167	4.2±0.058
M.excelsa sawdust	Control	4.2±0.120	6.7±0.120	2.1±0.060	3.3±0.060	2.7±0.058
	5%	5.3±0.058	7.4±0.116	2.6±0.030	3.7±0.058	2.9±0.116
	10%	5.7±0.058	7.6±0.116	3.0±0.029	3.8±0.033	3.2±0.120
	15%	6.1±0.058	8.6±0.145	3.1±0.029	4.3±0.101	3.4±0.058
	20%	6.4±0.116	8.8±0.033	3.1±0.088	4.5±0.017	3.8±0.033

Each value is an average mean of three replicates ±(SE = standard mean error of mean)

3. Results and Discussion

3.1Growth on different Agricultural wastes

All the agricultural wastes employed in these studies significantly supported the growth of *P. ostreatus* ($P<0.05$) .As shown on Table 1, the largest average length of pileus (8.00cm) was obtained on cotton waste (CW) with 20% rice bran (RB) additive.This was closely followed by rice straw (RS) (7.9cm) with 20% RB while the smallest length of pileus (4.20cm) was obtained on sawdust of *M. excelsa* without RB. Similar effect of different agricultural wastes on the growth of Lentinus *subnudus* were reported by Gbolagade *et al*., (2006a). The longest average stipe length, (8.80cm) was obtained on *M. excelsa* sawdust +20% RB and the smallest length, 2.90cm was obtained on RS without RB. The difference in the growth pattern of these substrates may be related to different chemical compositions of these agro-industrial wastes (Jonathan *et al*., 2012a). The sizes of the mushroom stipe and pileus may be affected by the substrates types. Fasidi *et al*., (2008), reported that very good mushroom pilei were produced on substrates with rice bran (containing 1.3% N). In addition, the size of the mushroom is dependent on substrates that were poor in cellulose, hemicellulose, lignin which constitute physical barrier and are difficult to be broken down without the presence of lignin degrading enzymes (Jonathan and Adeoyo, 2011). Good growths on agricultural substrates have also been linked with suitable nutrients and adequate environmental conditions (Gbolagade *et al.*, 2006b).

3.2: Nutritional compositions

Table 2 shows the proximate chemical analyses of *P. ostreatus* at different stages of development on *G. hirsutum* waste, *O. sativa* straw and *M. excelsa* sawdust. High moisture contents (94.34%) of fresh *P.osteratus* was recorded on cotton waste for young fruit bodies in the pileus and the least value (85.18%) was recorded for old fruit bodies in the stipe. This in line with the reports of Manzi *et al*, 1999; that moisture content of fresh *Pleurotus spp* ranged between 70 -95%. Very good crude protein content (28.0%) was obtained on cotton waste in young fruit bodies and the least value (16.33%) was recorded in stipe of over old fruit bodies. This results is line with the that of Fasidi *et al*,(2008). Fat content was generally low.8.72% was obtained for young fruit bodies in pileus while l4.80% was obtained in stipe of old sporophore(Table2).Cotton wastes are found to be supportive to growth and contained more nutrient values than saw dusts may be attributed to low lignin and hemicelluloses content of this substrate(Yildiz and Karakaplan 2003)

For rice straw (Table 2), the highest moisture content (93.13%) of *P. ostreatus* was observed on stipe of young fruit bodies while the lowest moisture content (84.39%) was recorded at over mature fruiting bodies. The highest crude protein content was 27.65% in the stipe while the lowest value (15.77%) was recorded on old pileus . The highest ash content (11.60%) was observed in old fruiting bodies while the lowest ash content (5.37%) was recorded with young fruiting bodies. Growth of *P. ostreatus* on sawdust of *M.excelsa* reveled that crude protein of 21.92% was obtained young pileus and low value(16.30%)was observed in the old stipe of this fungus. It could be observed from the results(Table1 and 2)that all the agricultural wastes used in this study generally supported good growth and nutritive values of *P. ostreatus*.This result is line with the suggestion of Fasidi *et al*.,(2008)that Pleurotus species could grow on wide varieties of agro-industrial wastes.

It was also observed from this study the *P.osteatus* grown on different wastes in these studies had high protein content.. Protein is one the most critical component contributing to the nutritional value of food (Gbolagade *et al*, 2006a and b). In this study, there are significant differences ($P < 0.05$) in the fat contents. Mushrooms are generally low in fat and have been suggested as ideal vegetable for the obesity(Kurtzman,2005). Jonathan *et al*, 2006 found 1.7% and 5.3% of fat and ash content of *Pleurotus florida* collected from the wild .These findings are lower than the value found for this study.The variations in values may be linked to the types of substrates and method of collection.

Table2: Proximate compositions of *P.osteatus* at different stages of sporophore development on agricultural wastes

Mushroom Part	Sporophore stage	Moisture %	Protein % (crude)	Fat %	Crude Fiber %	Ash %
Cotton waste	Young	91.00±0.20	21.56±0.01	8.40±0.01	16.70±0.01	6.090±0.003
Stipe	Matured	88.09±0.01	18.42±0.01	5.99±0.01	14.11±0.01	8.42±0.01
	Old	85.18±0.02	16.33±0.01	4.80±0.003	12.32±0.013	8.46±0.05
Cotton waste	Young	94.34±0.03	28.02±0.01	8.72±0.01	17.43±0.01	7.31±0.02
Pileus	Matured	88.37±0.02	25.92±0.03	6.80±0.01	14.79±0.01	8.02±0.01
	Old	85.87±0.02	26.35±0.02	4.94±0.01	12.78±0.002	9.03±0.00
Rice straw	Young	91.15±0.03	27.42±0.01	6.970±0.01	11.43±0.01	5.370±0.01
Stipe	Matured	88.02±0.01	18.32±0.01	5.220±0.01	9.330±0.01	7.350±0.02
	Old	84.39±0.10	16.07±0.02	3.930±0.02	7.430±0.02	8.500±0.00
Rice straw	Young	93.13±0.01	26.42±0.01	6.970±0.03	15.55±0.01	6.710±0.00
Pileus	Matured	88.22±0.01	27.65±0.02	5.720±0.01	10.16±0.01	9.450±0.01

	Old	84.45±0.20	15.77±0.02	4.090±0.00	8.540±0.02	11.60±0.02
Sawdust of *M. excels*	Young	90.83±0.01	21.92±0.01	6.930±0.01	14.43±0.01	6.060±0.01
Stipe	Matured	88.03±0.01	18 38±0.01	5.920±0.00	10.34±0.01	8.190±0.01
	Old	85.17±0.01	16.30±0.01	4.750±0.00	11.33±0.02	9.300±0.01
Sawdust of *M. excelsa*	Young	90.83±0.01	21.92±0.01	6.930±0.01	13.96±0.01	5.060±0.01
Pileus	Matured	88.32±0.01	19.52±0.01	6.420±0.01	12.12±0.01	8.20±0.02
	Old	85.60±0.01	17.32±0.01	4.870±0.01	12.42±0.03	12.00±0.00

Each value is a mean of three replicates ± S.E.M=S.E.M means Standard Error of the Mean

Table3: Vitamin composition of *P.ostreatus* cultivated on different agro-industrial wastes

Substrates/Vitamins (mg/100g)	***Vit. A***	***Vit.B1 (thiamine)***	***Vit.B2 (riboflavin)***	***Vit.B3 (niacin)***	***Vit.B5 (pantothenic acid)***	**Vit C (Ascorbic acid)**	***Vit.D***
Cotton wastes	0.00	0.92±0.28a	1.74±0.71a	2.13±0.30a	8.0±0.04a	3.65±0.17a	3.80±0.12b
Rice straw	0.00	0.67±1.30b	1.17±0.30c	2.75±0.68a	6.20±0.13b	3.52±0.42a	4.22±0.53a
Sawdust	0.00	0.35±1.00c	1.43±0.29b	1.72±0.50b	6.85±0.14b	3.27±0.47a	3.75±0.70b

Each treatment is a mean of three replicate with standard error of the mean (S.E.M ±).
Mean followed by the same letter are not significantly different at 5% level using DMRT.

Table 4. Biological efficiency of *P.ostreatus* on different substrates

Treatment	*Wet mushroom weight (g)*	*Dry mushroom weight (g)*	*Biological efficiency B.E (%)*
CW+CTRL	4.34±0.06a	0.56±0.09c	77.5
CW+5% RB	4.53±0.15a	1.04±0.01a	77.0
CW+10% RB	5.03±0.12c	1.56±0.01b	70.0
CW+15% RB	6.36±0.17b	1.64±0.01a	74.2
CW+20% RB	6.42±0.17a	1.68±0.02a	74.0
RS+CTRL	3.62±0.15d	0.23±0.03b	93.6
RS+5% RB	3.93±0.22d	0.28±0.04c	93.0
RS+10% RB	4.33±0.15ab	0.45±0.02c	89.6
RS+15% RB	4.80±0.06ac	1.08±0.06b	91.2
RS+20% RB	5.12±0.03b	1.23±0.10a	76.0
SD+CRTL	4.24±0.03b	0.05±0.01a	88.2
SD+5% RB	4.47±0.05c	0.98±0.02d	88.2
SD+10% RB	5.30±0.05c	1.34±0.18b	74.7
SD+15% RB	5.30±0.09b	1.40±0.09b	74.8
SD+20% RB	6.20±0.07d	1.51±0.20c	75.6

Each value represents the average mean of three different experiments with the Standard Error (S.E ±)
CW= cotton waste
RS= rice straw
SD= sawdust
RB= rice bran additive at varying percentage.
Mean followed by the same letters (s) within a treatment group are not significantly different at 5% and 1% level of probability using DMRT (Duncan Multiple Range Test).

3.3Vitamin composition:

Tabe 3 shows that *P.ostreatus* cultivated on various agricultural wastes in these studies were devoid of vitamin A, but possessed significant values of vitamin B complex (vitamin B1, B2, B3and B5) and appreciable amount of vitamin C and D.The most abundance vitamin(8.0mg/100g) was pantothenic acid on fruitbodies that were harvested on cotton wastes(Table3) However 6.85 and 6.20mg/100g of pantothenic acid were found on fruitbodies that were collected from sawdust and rice straw respectively. This result was similar to that observed by Kutzman (2005) for *Agricus bisporus* .This worker obtained 2.38 mg/100g for this white button mushroom. Pantothenic acid had been reported to function in nervous and hormonal balance (Çağlarırmak, 2007). The availability of vitamin B complex in the analyzed mushroom is not a surprise.this is because these vitamins especially niacin, riboflavin pantothenic acid and thiamine have been found in other mushrooms (Kurtzman,2005;Çağlarırmak,2007). Riboflavin have been found to stimulate healthy skin and vision, niacin aid in digestive and nervous system. It also aids pregnant women in the development of fetus (Bobek *et al.*, 1991;Jonathan *et al*,2011) . Vitamin C or ascorbic acid helps to fight against infection in red blood cell (Caglairmak, 2007). One cup of fresh *P.ostreatus* was recommended daily by Leigh(1994) for nornmal functioning of the body.

Cotton waste yielded more fruiting bodies than the other two substrates as shown on Table 4. This may be as a result of low lignin and hemicelluloses contents of cotton wastes as compared with wood wastes and rice straw(Gbolagade *et al*.,2006a). Use of rice bran supplement significantly increased the average weight of fruiting bodies. Rice bran is a vital basic nutrient for growth, being required for nucleic acid, protein, and chitin (in the case of fungi) synthesis (Fasidi *et al*,2008). Zadrazil and kuttzman, 1988, reported the yield of *pleurotus sp* can be boosted by the addition of nitrogenous supplements. It has been reported that urea,peptone and soya bean pomace were effective supplement to improve vegetative growth and fruitbody yield in mushrooms Gbolagade *et al*.,2006b; Fasidi *et al*,2008). Cotton waste supplemented with rice bran gave the best yield of *P.osteatus* in this study because this fungus may posses hydrolytic enzymes that were able to hydrolyse cellulose and hemicellulose into glucose and cellobiose (Albersheim, 1976; Keller, 1993). Leong (1978) reported that cotton waste was found to be better than rice straw and banana leaf substrate for *P. ostreatus* cultivation.

In conclusion, , cotton waste (*Gossypium hirsutum)* gave the best results in terms of yields, weights, nutritional value and chemical composition of *P. ostreatus* followed in order by *M.excelsa* sawdustand *O.sativa* straw. Rice bran was found to be an effective supplement that facilitated the growth of *P.ostreatus.* Rice bran should be incorporated as good additive with agricultural wastes to enhance growth.

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