

# Effect of growth supporting additives on the performance of Auricularia auricula on Mansonia altissima A. chev sawdust

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#### <u>Article history</u>

## <u>Abstract</u>

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## <u>Keywords</u>

Auricularia auricula Additives Proximate Composition Lignocelluloses composition Auricularia auricula (St. Aman's) Berk was cultivated on Mansonia altissima sawdust with various additives (Brewer's grain (BG), Corn chaff (CC), Oil palm fibre (OPF), Sorghum bicolor chaff (SC) and Wheat bran (WB) at different percentages (0%, 5%, 10% and 20%). The study was carried out to determine the effect of additives on the performance of the fungus on the substrate; M. altissima sawdust. The treated and untreated substrates with additives at different percentages were analyzed for lignocelluloses composition, macro element, C-N ratio and proximate composition. The result of this study showed that A. auricula reduced the lignocelluloses composition of M. altissima sawdust. The lignin content reduced from 7.97% (control) to 1.59% in 20%SC treated substrate. The macro elements (Ca, Mg, K, Na) compositions were low in all the treated substrate - additives combination. The least was recorded in Na (25.8 - 84.5ppm), Ca (2.04% in control and 0.50% in 20%SC). The proximate composition showed that the substrate had an average moisture content of 50% - 61%, low protein (4.85-0.60%), high carbohydrate and high ash contents compared to the control. The results of this study showed that A. auricula exhibited an increase in performance with increase in additives, with the most efficient at 20% for all the additives. It can also be concluded that wheat bran was the best out of the five additives used.

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## Introduction

Auricularia auricula (wood ear) mushroom is the fourth most important cultivated mushroom in the world (PeiSheng *et al.*, 2004). It has a gelatinous, rubbery texture with a unique jelly taste and the basidium is horizontally septated. This makes it to be different from other cultivated mushrooms. *Auricularia* spp. grows naturally on truck and roots of trees as well as on decaying wood materials (Iwalokun *et al.*, 2007). A wide range of diverse lignocelluloses substrates are used for cultivation of mushroom and their rapidity of growth makes their cultivation possible in different parts of the world.

Mushrooms are grown on lignocellulosic materials, which are obtained in large quantities as waste residues in agriculture and forestry and they often constitute an abundant but underutilized source of renewable biomass (Hartley *et al.*, 1987). Large amount of this renewable biomass are potential source of bio fuels, chemicals, cheap energy source for fermentation, improved animal feeds and human nutrients (Howard *et al.*, 2003). The chemical properties of the components of lignocelluloses

make them a substrate of enormous biotechnological value (Malherbe and Coete, 2003). Lignocellulosic materials are generally low in protein content, insufficient for the cultivation of mushrooms that requires nitrogen, potassium, etc. Since C: N ratio plays an important role in spawn running and the growth of fruiting body, the addition of organic nitrogen supplements is an important factor for growth and growth yield of mushrooms (Mane et al., 2007). Additives are normally rich in Nitrogen and Carbon or both. Nitrogenous supplements include ammonium salts, urea, yeast powder, wheat bran, soybean meal, sesame and peanut while carbohydrate nutrients supplements such as molasses, corn chaff, rice bran, oat meal and sugar were found to accelerate mycelia growth of Lentinus edodes and Auricularia spp. (Han et al., 1981; Quimio, 1981). Inclusion of additives to substrates with different levels of carbonates and nitrogen- based additives have been shown to enhance mushroom production (Isikhuemhen et al., 1999).

In Nigeria, various researchers had studied growth performance of *Pleurotus* (Isikhuemhen *et al.*, 1999; Okhuoya *et al.*, 2000; Joshua and Agina, 2002) and *Volvariella* (Fasidi and Kadiri, 1993; Kuforiji and Fasidi, 2006) and *Lentinus* spp. (Ayodele and Akpaja, 2007; Okhuoya *et al.*, 2005) but there has been little or no attempt on *Auricularia* spp. commonly found in Nigeria and eaten by various tribes in one form or the other.

The *Auricularia* reported in this work is a Ghanaian mushroom cultivated under Nigeria environmental conditions. We have started the domestication studies of this mushroom in Nigeria (Lawal *et al.*, 2011). This work is aimed at evaluating the effect of different additives at different levels on performance of *Auricularia auricula* on *Mansonia alltissima* sawdust.

# **Materials and Methods**

#### Sample collection

*Mansonia alltissima* (A.chev) sawdust collected from Sango sawmill market in Ibadan, Nigeria was used as substrate. Five organic supplements were used as additives at three different levels of inclusion. The additives used were brewer's grain (BG) from Nigeria breweries PLC Ibadan; Corn chaff (CC) and Sorghum chaff (SC) collected from Agbowo in Ibadan(suburb of University of Ibadan), wheat bran obtained from Kara , Bodija market, Ibadan, Oyo State and oil palm fibre (OPF) was collected from a local oil mill at Akungba, Ondo State.

#### Spawn preparation

The pure culture of the fungus MBFBL strain 266 was obtained from the Mushroom Biology and Food Biotechnology Laboratory, North Carolina Agricultural and Technical State University, Greensboro, USA.

Spawn was prepared with intact sorghum grains. Five hundred grams of the grains were weighed, mixed with 1% calcium carbonate (CaCO<sub>3</sub>) and 10% wheat bran.(Lawal *et al.*, 2011).The grains were then filled into micro-pore fitted polypropylene bags (Unicorn bags, USA) and sterilized at 121°C at 15 lbs pressure for 15 minutes. The bags were inoculated with a pure culture of *A. auricula* under aseptic conditions and incubated at room temperature ( $28\pm2^{\circ}C$ ) for mycelial growth.

#### Substrate preparation.

Five hundred grams (500g) of the dried *Mansonia* altissima sawdust was soaked in water for two hours and excess water was squeezed out using muslin cloth. 1% CaCO<sub>3</sub> was added to the sawdust along with the various additives at 0% (control), 5%, 10% and 20%. The additives and lime were thoroughly

mixed with the sawdust and loaded into well labeled polypropylene bags (Lawal *et al.*, 2011). The bags were steamed in a drum for four hours and left in the drum until the following day. The bags were then inoculated with *A. auricula* spawn under aseptic conditions and incubated at room temperature  $(28\pm2^{\circ}C)$  in the dark for mycelia growth in the mushroom house, Department of Botany, University of Ibadan. At the sight of primordial formation, the bags were opened, soaked in water and exposed to light for fructification. The treated substrates were the sawdust mixed with additives inoculated with the fungus (*A. auricula*) while the untreated substrates were sawdust mixed with additives without *A. auricula* inoculation.

#### Mineral determination

One gram of each sample (Untreated and treated(spent) sawdust-additive substrates) was ashed in a furnace and washed with 2 ml concentrated nitric acid ( $HNO_3$ ) and the filtrate was made up to 50 ml with distilled water. The mineral contents were determined by automated atomic absorption spectrophotometry and photometry methods according to the methods of AOAC. (2003).

#### Lignocelluloses composition

These were estimated using the methods of Van Soest, 1963, Van Soest and Wine 1969; Goering and VanSoest, 1970. These methods involved the estimation of cell wall as neutral detergent fibre and lignin as acid detergent and lignocelluloses as acid detergent fibre.

#### Protein content

The protein content was determined using the Micro-Kjeldal automated method. 0.2 grams of the dried fruit bodies was weighed into a digestion tube and 15ml of conc.  $H_2SO_4$  and 7 Macro kjeldhal tablets was added into the digestion per set at 410°C. Digestion was done at 410°C for 45minutes until when there was a clear solution.

#### Ash content

Two grams of powdered mushroom sample was reduced to ash in Gallenkamp furnace in a crucible that had been ignited, cooled and weighed at 550°C for 6 hours (Campbell *et al.*, 1968).

# Crude fibre content

This was determined using the sulphuric acid method. 0.1 gram of powdered sample was weighed into a beaker and 200 ml of hot 1.25% H<sub>2</sub>SO<sub>4</sub> was added. The beaker was boiled and refluxed for 30

	LI	GNIN	CELL	ULOSE	HEMICELLULOSE		
ADDITIVES	TREATED	UNTREATED	TREATED	UNTREATED	TREATED	UNTREATED	
CONTROL	7.973 <sup>e</sup>	8.863 <sup>f</sup>	12.80 <sup>k</sup>	11.82 <sup>l</sup>	4.910 <sup>f</sup>	4.977 <sup>j</sup>	
5% BG	3.870ª	5.993ª	6.920ª	10.10ª	3.207ª	3.547ª	
10% BG	2.370 <sup>f</sup>	3.117 <sup>b</sup>	6.477 <sup>9</sup>	8.810 <sup>e</sup>	3.220°	3.123 <sup>9</sup>	
20% BG	1.740 <sup>f</sup>	1.923°	6.523 <sup>i</sup>	10.43 <sup>g</sup>	3.063°	2.547°	
5% CC	4.827 <sup>b</sup>	4.777ª	5. <b>7</b> 80ª	8.830 <sup>b</sup>	2.187ª	2.940 <sup>b</sup>	
10% CC	2.500 *	4.910 <sup>d</sup>	5.387 *	9.160ª	3.200°	1.943 <sup>h</sup>	
20% CC	1.717 *	2.070 °	8.967 <sup>d</sup>	7.420 <sup>n</sup>	1.913 <sup>e</sup>	1.693°	
5% OPF	3.927 <sup>e</sup>	5.773ª	4.910ª	9.440ª	1.843ª	2.140 °	
10% OPF	2.370 <sup>†</sup>	2.367 °	7.197 °	7.617 '	1.770 °	1.677 <sup>t</sup>	
20% OPF	1.763 *	2.080 °	5.923 °	6.730 <sup>†</sup>	1.843 <sup>e</sup>	1.597°	
5% SC	2.037 <sup>f</sup>	3.000 <sup>b</sup>	4.300ª	8.763 °	1.887 <sup>d</sup>	1.720 <sup>d</sup>	
10% SC	2.267 <sup>f</sup>	2.783°	4.027 <sup>b</sup>	6.180 <sup>i</sup>	1.150 °	1.040 <sup>f</sup>	
20% SC	1.590 °	1.743 °	5.587 <sup>j</sup>	8.193 <sup>f</sup>	1.137 <sup>e</sup>	1.043°	
5% WB	2.130 <sup>f</sup>	3.040 <sup>b</sup>	4.690ª	6.247°	1.077 <sup>b</sup>	1.127 <sup>i</sup>	
10% WB	1.817 <sup>f</sup>	2.493 °	3.910 <sup>h</sup>	6.160 <sup>k</sup>	1.160 °	1.070 <sup>f</sup>	
20% WB	1.800 <sup>f</sup>	1.863 °	4.667 <sup>€</sup>	6.337 <sup>f</sup>	1.060 <sup>e</sup>	1.130 <sup>●</sup>	

Table 1. Lignocellulose contents of M. altissima sawdust with additives inoculated with A. auricula

Each figure is a mean of 3 readings. Values in the same column followed by the same letters are not significantly different at  $P \le 0.05$  according to Tukey's multiple test ( $P \le 0.05$ )

minutes on a digestion apparatus with preheated plates. The resulting suspension was filtered through Whatman No 1 paper by gravity and the residue on the paper washed with water until the filtrate was neutral.

The residue was then dried in a previously ignited, cooled crucible at 100°C overnight. The crucible was cooled in a desiccator and weighed (weight A). The content was then ashed in a furnace at 600°C for 6 hours and the crucible cooled again in a desiccator (weight B).

## Moisture content

Fresh samples of mushrooms were harvested and weighed. The samples were then dried in the oven at 80°C for two days. The percentage difference in fresh and dry weight was taken as the moisture content according to the method of Campbell *et al.* (1968).

## Ethanol- soluble sugar content

One gram of powdered sample was extracted overnight with 25 ml of 80% ethanol. The extract was filtered out and the filtrate made up to 100 ml with distilled water. The quantity of ethanol-soluble sugar in the extract was determined using phenol sulphuric acid method of Dubois *et al.* (1956).

#### **Results and Discussion**

#### Lignocellulose composition

Table 1 shows the lignocelluloses composition of *M. altissima* with additives treated and untreated with A. auricula. The lignin content in the treated substrates recorded a significant reduction in percentage with increase in percentage of additives. Also the treated substrates recorded a reduction in lignin compared to the untreated. Over all, the least lignin content (1.59%) was recorded for sawdust + 20% additives. The reduction in lignin content (7.97% in control -1.59% for sawdust treated with 20%SC) showed that A. auricula was able to break lignin barrier and other complex polysaccharides present in the substrate. The lignin content ranges between 1.59% for 20% SC to 7.97% for the control experiment for treated substrates while the untreated recorded a reduction from 8.86% in the control to 1.74% for 20% SC. The reduction in the lignin content of the treated substrate revealed that there was significant difference in all the substrate additive combinations as shown in Table 1.

The reduction in lignin content recorded is similar to the findings of Adenipekun and Fasidi (2005) in rice straw inoculated with *P. tuberregium* and *L. subnudus*. Zadrazil (2000) observed that biological delignification using lignin degrading fungi increase

	CALCIUM (%)		POTASSIUM (%)		MAGNE	ESIUM (%)	SODIUM (PPM)		
ADDITIVES	TREATED	UNTREATED	TREATED	UNTREATED	TREATED	UNTREATED	TREATED	UNTREATED	
CONTROL	2.040 <sup>d</sup>	2.160 <sup>⊳</sup>	0.383°	0.395 °	0.913 <sup>d</sup>	0.923 <sup>e</sup>	84.447 <sup>b</sup>	86.653°	
5%BG	0.650°	1.430ª	0.145ª	0.365 ª	0.310ª	0.563ª	26.553°	63.890ª	
10%BG	0.587 <sup>b</sup>	1.057ª	0.120 <sup>b</sup>	0.194 ª	0.310 <sup>b</sup>	0.500 <sup>b</sup>	28.440ª	53.040 <sup>b</sup>	
20%BG	0.520 <sup>b</sup>	0.757 <sup>b</sup>	0.109 <sup>b</sup>	0.216 ª	0.267ª	0.543ª	26.117ª	51.907 <sup>b</sup>	
5%CC	0.693ª	1.420ª	0.122ª	0.205 ª	0.327ª	0.61 <b>7</b> ª	34.287ª	61.003ª	
10%CC	0.630 <sup>b</sup>	1.043ª	0.116 <sup>⊳</sup>	0.192 ª	0.317 <sup>b</sup>	0.457 <sup>b</sup>	28.917ª	59.537°	
20%CC	0.515 <sup>⊳</sup>	1.080ª	0.113 <sup>b</sup>	0.230 ª	0.300 <sup>b</sup>	0.423°	25.867ª	62.237°	
5%OPF	0.640ª	1.330ª	0.617°	0.222 ª	0.293ª	0.747 <sup>d</sup>	27.523ª	63.080 ª	
10%OPF	0.710 <sup>b</sup>	1.020ª	0.126 <sup>⊳</sup>	0.243 ª	0.360 <sup>b</sup>	0.557°	27.047ª	63.660 ª	
20%OPF	0.580 <sup>b</sup>	1.040ª	0.121 <sup>⊳</sup>	0.215 °	0.363 <sup>b</sup>	0.497 <sup>b</sup>	26.353ª	60.627 ª	
5%SC	0.683ª	1.313ª	0.119 <sup>b</sup>	0.428 ª	0.320ª	0.593ª	29.570°	58.590 ª	
10%SC	0.510 <sup>⊳</sup>	1.027ª	0.127 <sup>b</sup>	0.209 ª	0.290ª	0.447 <sup>a</sup>	33.647ª	50.937 <sup>b</sup>	
20%SC	0.500 <sup>b</sup>	1.027ª	0.122 <sup>⊳</sup>	0.216 ª	0.277ª	0.487 <sup>⊳</sup>	27.697ª	53.563 <sup>b</sup>	
5%WB	1.123°	1.367ª	0.119ª	0.228 ª	0.155°	0.637ª	26.243ª	60.733ª	
10%WB	0.703 <sup>b</sup>	0.987ª	0.116 <sup>⊳</sup>	0.216 ª	0.300 <sup>b</sup>	0.527ª	29.117ª	52.820 <sup>b</sup>	
20%WB	0.583 <sup>b</sup>	1.193ª	0.114 <sup>b</sup>	0.252 °	0.297ª	0.527 <sup>b</sup>	29.180°	59.103 <sup>b</sup>	

Table 2. Macroelement Composition of M. altissima with additives treated and untreated with A.auricula

Each figure is a mean of 3 readings. Values in the same column followed by the same letters are not significantly different at P $\leq$  0.05 according to Tukey's multiple test (P $\leq$  0.05)

the lignin content in plant material used as feed correlates with the decrease in digestibility for rumen microorganism. The highest percentage cellulose was found in the control (12.80%) which was significantly different from 3.91%, the least cellulose content was recorded for sawdust + 20% SC. The percentage hemicellulose content reduced significantly ( $P \le$ (0.05) in the treated substrates from 4.91% in control to 1.06% in sawdust + 20% WB as shown in Table1. This report corroborates the finding of Lu and Tang, (2006) who reported the ability of A. polytricha to hydrolyze cellulose and hemicellulose due to the substantial amounts of cellulose and hemicellulose enzymes produced. The ability of this fungus to reduce the polysaccharide complex makes their spent substrate a potential source of organic fertilizer.

#### Macroelements composition

The macro elements compositions of *M. altissima* with additives inoculated with *A. auricula* is shown in Table 2. The calcium content in the treated substrates was reduced significantly compared to the untreated substrates. The calcium content reduced with increase in percentage of additives from 5% to 20% for all the additives used. Calcium is the most abundant element in the substrates additives composition as shown in Table 2. This is likely to have resulted from the addition of CaCO<sub>3</sub> to the substrate during the preparatory stage. The potassium content in the treated substrates ranges between 0.109% (20% BG) to 0.145% (5%BG).

Sodium was the least of the entire macro element tested; this indicates that the substrate contained little or no sodium. This low content of mineral recorded suggested that the mushroom was able to mineralize the substrate and take up mineral element into its tissues thereby increasing its dietary quality (Tyler, 1982). This report is in line with the findings of Rudawaska and Leski (2005) who reported that macro elements concentrates in the fruit bodies of mushroom than in the substrates where they were collected.

Malinowska et al. (2004) also reported a wide range of macro elements concentrations in the fruit bodies of mushroom (wild and cultivated) irrespective of their geographical origin or how the fungi acquire nutrients (saprotroph, nectotroph or biotrophs). Substrate composition is an important factor besides the great differences that exist in uptake of individual trace elements by the fruit body of mushrooms reported by Kalac and Svobadan(2000), Nikkarinen and Mertanen (2004). This implies that mushrooms have a specialized mechanism for accumulation of nutrients into their fruit bodies. The concentrations of elements in fruiting bodies of mushrooms are generally species dependent. However, there is clear tendency towards higher macro element nutrient accumulation in the caps than in stripes.

The carbon content in the treated substrates reduced significantly with increase in percentage of additives compared to untreated substrates as shown on Table 3. The carbon content ranged between 0.94% (10%WB) to 8.64% in the control for treated substrates. The highest nitrogen content was found in the control (0.80%) followed by 0.18% in 5% WB; these values were found to be significantly different (P≤0.05). The reduction in carbon and nitrogen content reported in this work is similar with the report

	CARBON		NITE	ROGEN	C:N RATIO		
ADDITIVES	TREATED	UNTREATED	TREATED	UNTREATED	TREATED	UNTREATED	
CONTROL	8.64 <sup>d</sup>	9.23 <sup>i</sup>	0.80 <sup>b</sup>	0.80 <sup>k</sup>	10.79 <sup>b</sup>	11.48ª	
5%BG	1.19ª	3.85 °	0.12 ª	0.39ª	10.37ª	11.50ª	
10%BG	1.15 <sup> b</sup>	3.92 <sup>d</sup>	0.12 ª	0.40 °	9.83ª	10.14 <sup>b</sup>	
20%BG	1.09 <sup>b</sup>	2.27 °	0.11 ª	0.27 <sup>f</sup>	9.81ª	11.01ª	
5%CC	1.37 ª	4.69 ª	0.14 ª	0.39ª	10.54 ª	10.72ª	
10%CC	1.06 <sup>b</sup>	3.14°	0.10 ª	0.25°	10.24 ª	11.27 ª	
20%CC	0.98 •	3.31°	0.11 ª	0.31 <sup>ª</sup>	9.89ª	10.94 ª	
5%OPF	1.26 ª	5.36 <sup>b</sup>	0.12 ª	0.54 <sup>9</sup>	10.54 ª	11.74 °	
10%OPF	1.03 <sup>b</sup>	3.05 <sup>g</sup>	0.12 ª	0.29 <sup>b</sup>	9.37ª	10.07 <sup>b</sup>	
20%OPF	1.34 <sup>b</sup>	2.91 °	0.13 ª	0.28 <sup>d</sup>	10.35 ª	11.28 ª	
5%SC	1.24 ª	4.03 <sup>f</sup>	0.12 ª	0.31 °	9.71ª	11.29 ª	
10%SC	1.28 <sup>b</sup>	4.05 <sup>d</sup>	0.13 ª	0.37 <sup>b</sup>	10.27 ª	11.43 ª	
20%SC	1.13 <sup>b</sup>	3.06 <sup>g</sup>	0.11 ª	0.31 <sup>d</sup>	10.76 ª	12.01 °	
5%WB	2.63 °	6.31 <sup>h</sup>	0.18 ª	0.51 <sup>i</sup>	10.38 ª	12.18 °	
10%WB	0.94 <sup>b</sup>	4.14 <sup>d</sup>	0.09 ª	0.12 <sup>i</sup>	9.79ª	11.09 ª	
20%WB	1.27 <sup>b</sup>	2.51 °	0.12 ª	0.25 <sup>h</sup>	10.46 ª	10.9ª	

Table 3. Carbon –nitrogen ratio composition of *M. altissima* with additives treated and untreated with *A. auricula* 

Each figure is a mean of 3 readings. Values in the same column followed by the same letters are not significantly different at  $P \le 0.05$  according to Tukey's multiple test ( $P \le 0.05$ )

	MOISTURE		PROTEIN		Cł	CHO		FAT		CRUDE		ASH	
ADDITIVES	Trt	Untrt	Trt	Untrt	Trt	Untrt	Trt	Untrt	Trt	Untrt	Trt	Untrt	
CONTROL	51.39ª	52.45 <sup>b</sup>	4.85 <sup>d</sup>	4.95 <sup>n</sup>	12.32 <sup>d</sup>	8.69 <sup>9</sup>	3.37°	3.66 <sup>h</sup>	4.03 <sup>g</sup>	4.39 <sup>g</sup>	22.01ª	32.86 <sup>b</sup>	
5%BG	61.11ª	50.79ª	0.72ª	2.27ª	9.17ª	9.67 <sup>de</sup>	1.09ª	3.12ª	0.79ª	2.16ª	19.03 <sup>bc</sup>	30.99°	
10%BG	58.75ª	58.90 ª	0.75 <sup>b</sup>	2.35 <sup>d</sup>	10.08°	12.39 <sup>b</sup>	1.03 <sup>b</sup>	2.16 <sup>b</sup>	0.67 <sup>b</sup>	1.82 <sup>b</sup>	17.02 <sup>bc</sup>	21.35°	
20%BG	54.25ª	53.11 <sup>®</sup>	0.69 <sup>b</sup>	1.59 <sup>₫</sup>	10.21°	12.08 <sup>b</sup>	1.05ª	1.41°	0.51°	1.33°	16.01 <sup>bc</sup>	21.49 <sup>jk</sup>	
5%CC	60.18ª	50.63 ª	0.78ª	2.69 <sup>b</sup>	10.76°	10.42 <sup>b</sup>	1.10ª	3.47 <sup>d</sup>	0.78ª	2.17ª	16.93 <sup>bc</sup>	33.00 <sup>b</sup>	
10%CC	61.73ª	59.57 ª	0.66 <sup>b</sup>	1.66 <sup>e</sup>	9.17ª	9.22 <sup>de f</sup>	1.18ª	2.21 <sup>b</sup>	0.64 <sup>d</sup>	1.89 <sup>b</sup>	19.24 <sup>bc</sup>	29.38°	
20%CC	57.77ª	53.67 <sup>b</sup>	0.63 <sup>b</sup>	1.98'	12.14 <sup>b</sup>	8.26 <sup>g</sup>	0.97°	1.42°	0.48 <sup>e</sup>	1.25°	13.47°	21.18 <sup>ki</sup>	
5%OPF	61.05ª	55.95ª	0.75ª	3.08 <sup>g</sup>	10.68°	14.53ª	1.11ª	2.94 <sup>e</sup>	0.79ª	2.18ª	19.57 <sup>bc</sup>	25.50 <sup>i</sup>	
10%OPF	59.60ª	60.26 ª	0.70 <sup>b</sup>	1.93 <sup>h</sup>	11.01°	10.98°	1.13ª	2.27 <sup>b</sup>	0.55'	1.65₫	20.92 <sup>bc</sup>	26.36 <sup>g</sup>	
20%OPF	55.12ª	53.86 <sup>b</sup>	0.82 ª	1.64 <sup>i</sup>	9.98 °	9.52 <sup>ef</sup>	1.11ª	1.48°	0.49 <sup>e</sup>	1.03°	19.64 <sup>bc</sup>	18.60 <sup>n</sup>	
5%SC	55.99ª	51.21 <sup>b</sup>	0.76ª	2.17 <sup>j</sup>	9.70°	8.82 <sup>fg</sup>	1.06ª	3.18 <sup>f</sup>	0.73ª	1.95ª	22.25 <sup>b</sup>	29.91 <sup>d</sup>	
10%SC	56.69ª	52.07 <sup>b</sup>	0.76 <sup>b</sup>	2.18 <sup>k</sup>	10.04°	10.02 <sup>d</sup>	1.06ª	2.25 <sup>b</sup>	0.62 <sup>d</sup>	2.08 <sup>e</sup>	15.77 <sup>bc</sup>	57.36ª	
20%SC	58.49ª	51.99 <sup>b</sup>	0.66 <sup>b</sup>	1.61 <sup>i</sup>	10.56°	11.18°	1.02 <sup>b</sup>	1.31 <sup>g</sup>	0.51°	1.34°	16.12 <sup>bc</sup>	25.70 <sup>h</sup>	
5%WB	58.33ª	50.80 <sup>b</sup>	1.59°	3.22	10.14°	10.70°	1.25ª	3.04 <sup>e</sup>	0.77 <sup>a</sup>	2.90'	21.89 <sup>b</sup>	21.70	
10%WB	50.03ª	56.40 ª	0.60 <sup>b</sup>	2.18 <sup>m</sup>	9.46 °	11.06°	1.08ª	1.54°	0.61 <sup>f</sup>	1.93 <sup>⊳</sup>	20.85 <sup>bc</sup>	27.06 <sup>f</sup>	
20%WB	52.69ª	54.46 ⁵	0.75 <sup>⊳</sup>	1.43'	10.39°	9.907 <sup>de</sup>	1.11 <sup>a</sup>	1.34 <sup>g</sup>	0.48 <sup>e</sup>	1.04°	<b>15.86</b> ⁵°	19.93 <sup>m</sup>	

Table 4. Proximate composition of M. altissima with additives treated and untreated with A. auricula

Each figure is a mean of 3 readings. Values in the same column followed by the same letters are not significantly different at  $P \le 0.05$  according to Tukey's multiple test ( $P \le 0.05$ ).

of Nageswaran *et al.* (2003), where they reported a reduction in carbon and nitrogen content of paddy straw after harvesting oyster mushrooms compared with the pre-harvest samples. The presence of nitrogen in the spent substrate makes it a good source of soil conditioner / fertilizer. Oei (2003) reported that

all mushroom spent substrates could be used as soil conditioner or fertilizers, but it is sometimes better to compost the spent substrate before its application to the soil, otherwise it might draw valuable nitrogen from the soil.

# Proximate composition

The proximate composition of M. altissima sawdust is shown in Table 4. The Protein content ranged from 0.60% to 4.85% in the treated substrates, showing that the protein content decreases in the substrates with increase in the percentage of additives. This result differs from the findings of Belewu (2001) who reported that the protein content of treated substrates was significantly higher than untreated substrates due to the addition of fungal protein. The crude protein content of mushroom treated substrate increased with degradation process while the crude fibre content in the treated substrates decreased with an increase in percentage of additives compared with the untreated substrates. The relatively lower crude fibre contents might be due to lignin and cellulose degradation by the mushroom. This degradation would increase the digestibility of this residue (spent mushroom substrate) due to the presence of mycelium (Sueli, 2002).

Therefore the residue (mushroom treated substrates) can be used as nutritional complement in ration for animal's feeds.

# Conclusion

The addition of additives to sawdust; Brewer's Grain (BG), Corn Chaff (CC), Wheat Bran (WB), Oil palm fruit fibre (OPFF) and sorghum chaff (SC) significantly increased the mycelia extension, density and yield of *A. auricula*. The optimum additives percentage varies with additives used. The ability of *A. auricula* to degrade the lignocelluloses complex and the presence of mineral elements rendered the spent compost as a good potential organic fertilizer. The report of this work recommends the use of *M. altissima* sawdust with Brewer's Grain (BG), Corn Chaff (CC) and Wheat Bran (WB), for the cultivation of this fungus. However wheat bran is the best out of the five additives used.

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