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Degradation of lignocellulosic substrates by Pleurotus ostreatus and Lentinus squarrosulus

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ABSTRACT

Lignocellulosic substrates are wastes in the environment whose reducing sugars are not readily available for use. Biological pretreatment is the use of microorganisms and/or their metabolites to break down substrates to obtain simple sugars which is also cheap compared with other pretreatment techniques. This work is aimed at degrading lignocellulosic substrates with higher mushrooms to obtain simple sugars that could be used as raw materials for other industrial processes. The two mushrooms [*Pleurotus ostreatus* (PO) and *Lentinus squarrosulus* (LS)] with the ability to produce cellulase, xylanase, and lignase were used for degradation of lignocellulosic substrates [groundnut shell (GS), maize cob (MC), maize straw (MS), rice straw (RS) and sugarcane bagasse (SB)]. The residual extractives, cellulose, hemicellulose, lignin, and reducing sugar contents were determined every 7 days. Least extractives (1.12 %), hemicellulose (15.09 %), lignin (17.60 %), and cellulose (5.60 %) were recorded in PO-degraded MS, POLS-degraded GS, LS-degraded GS, and PO-degraded MS at 28, 35, 49 and 42 days of degradation, respectively. The highest reducing sugar contents (mg/g) obtained in GS (11.83), MS (27.03), SB (28.70), and RS (37.96) were recorded when degraded by PO for 49, 14, 7, and 49 days, respectively while that of MC (13.32) was recorded when degraded by LS for 42 days. Reducing sugar obtained was higher from sole degradation with PO compared with LS and POLS. Degraded MS, RS, and SB had better yield of reducing sugar than GS and MC. The amount of reducing sugar released varied with substrates, organisms, and degradation time.

Contribution/Originality: This study showed that the amount of reducing sugar released from different lignocellulosic materials varied with substrates, organisms and degradation time, and the reducing sugar was higher from sole degradation with *Pleurotus ostreatus* compared with *Lentinus squarrosulus* and co-cultured strains.

1. INTRODUCTION

Second-generation biofuels are produced from non-edible agricultural wastes however, reducing sugars needed for the generation of biofuel in these wastes are not readily released from them [1-3]. Therefore, it is necessary to pretreat non-edible agricultural wastes to obtain simple sugars needed for the production of biofuel. Non-edible agricultural wastes are lignocellulosic substrates that contain mainly cellulose, hemicellulose, and lignin [4]. The cellulose and hemicellulose when broken down contain fermentable sugar but lignin does not contain any sugar [5, 6]. Biological, chemical and physical, and physicochemical methods of pretreatment are available [7, 8]. The biological pretreatment method is the cheapest among different pretreatment techniques and it is also environmentally friendly [9]. Organisms or their metabolites are used to break down lignocellulosic substrates in the biological pretreatment method. Lignocelluloses are broken down by soft rot, brown rot, and white rot fungi [10]. There are two kinds of white rot fungi such as selective and non-selective. Hemicellulose and lignin portions of lignocellulose are degraded by selective white rot fungi while the cellulose part is essentially not degraded but all parts of lignocellulose are equally degraded by non-selective white rot fungi [10-12]. Some of the white rot fungi that have been utilized to degrade lignocellulose are *Obba rivulosa Ceriporia lacerate*, *Ceriporiopsis subvermispora*, *Cyathus stercolerus*, *Pycnoporus cinnarbariunus*, *Lentinus squarrosulus*, *Pleurotus ostreatus*, *Pleurotus tuber-regium*, *Gelatoporia subvermispora* and *Phanerochaete chrysoporium* [1-3, 9, 13, 14].

Lignocellulosic wastes in the environment could include groundnut shell, maize straw, rice straw, sugar cane bagasse, maize cob, and others. Utilization of these non-edible wastes will reduce waste in the environment. Degradation of these lignocellulosic substrates with fungi and obtaining a good yield of reducing sugar required for fermentation of biofuel and other industrial processes would make this technique economically viable. This work aimed at degrading lignocellulosic substrates with fungi to obtain fermentable sugars.

2. MATERIALS AND METHODS

2.1. Collection of Lignocellulosic Wastes

All lignocellulosic biomass were collected from Oyo state (8.1196° N, 3.4196° E). Groundnut shell was collected from Saki, maize cob from Ajegunle, Oyo town while maize straw was collected from Okunlola's farm, Ilora. Sugar cane bagasse was obtained from Akunlemu, Oyo town and rice straw was collected from the International Institute of Tropical Agriculture (IITA), Ibadan. All the samples were dried and milled with a milling machine in Oyo town and taken to the Laboratory of the Department of Biological Sciences, Ajayi Crowther University, Oyo town.

2.2. Collection of White Rot Fungi

Two white rot fungi (*Lentinus squarrosulus* and *Pleurotus ostreatus*) with the potential of producing cellulases, hemicellulases, and lignin-modifying enzymes were collected from the Department of Botany, University of Ibadan, Ibadan.

2.3. Pretreatment of Lignocellulose with White Rot Fungi

Five lignocellulosic wastes (maize cob, maize straw, rice straw, groundnut shell, and sugar cane bagasse) were pretreated with *Pleurotus ostreatus* and *Lentinus squarrosulus* singly and in consortium. One hundred (100) grams of each lignocellulosic material were weighed into separate glass jars and 300 mL of distilled water was added and mixed. They were sterilized at 121 °C and 1.05kg cm^{-2} for 15 minutes and allowed to cool. Each cooled biomass was inoculated separately with eight agar plugs (7mm in diameter) of *Pleurotus ostreatus* and *Lentinus squarrosulus* separately and consortium (4 agar plugs each of *Pleurotus ostreatus* and *Lentinus squarrosulus*) [2]. They were incubated at 28 ±2°C for 70 days. Samples were taken from degrading substrates every 7 days and were analyzed for cellulose, hemicellulose, lignin, extractives, and reducing sugar contents.

2.4. Determination of Extractives, Hemicellulose, Lignin, and Cellulose

The extractive in each biomass was determined using solvent extraction. Sixty millilitres of acetone was added to a dried biomass sample (1 g). The temperature was maintained at 56 ℃ for 2 hours. The samples were then dried at 105 ℃ until they reached a constant weight. The amount of extractives in the biomass is the weight difference between the sample before and after extraction [15]. Extractive-free dried biomass (0.4 g) was added to four millilitres of 0.5 mol/L NaOH, and the mixture was maintained at 80 ℃ for three and a half hours. The sample was washed with distilled water until the solution's pH reached 7, and it was then dried in a hot air oven at 105 ℃ until a constant weight was observed. The amount of hemicellulose is the weight difference between the sample's initial and final weights [15]. Extractive-free dried biomass (0.4g) was mixed with 12 millilitres (98% sulphuric acid) and left at room temperature for 24 hours. The sample was diluted with 80 mL of distilled water after 24 hours, and it was then heated for 1 hour at 100 °C. The mixture was allowed to cool, filtered, and the residue was washed with distilled water until there were no sulphate ions left in the filtrate. The residue was dried until it reached a constant weight. The lignin content is the weight of the dried residue [15].

The difference was used to compute the amount of cellulose, with the assumption that each biomass consisted solely of cellulose, lignin, hemicellulose, and extractive [15].

Weight of the total biomass - (lignin + hemicellulose + extractive) content equals the content of cellulose

2.5. Determination of Reducing Sugar Content

Extraction of reducing sugar from the degraded substrate was done by adding 1 g of the degraded substrate to 20 mL of distilled water and shaking every 15 minutes for 2 hours. It was filtered, and the obtained filtrate was utilized to determine the amount of reducing sugar. The Dinitrosalicylic acid method was used to determine the amount of reducing sugar [16].

2.6. Statistical Analysis

The level of significance was set at $P \leq 0.05$ for the analysis of variance, which was performed on the experimental data to estimate the means, using SPSS version 23.

3. RESULTS

The component (extractive, hemicellulose, lignin, and cellulose) of groundnut shell pretreated with *Pleurotus ostreatus* and *Lentinus squarrosulus* is shown in Table 1. Extractives of groundnut shell degraded by *Pleurotus ostreatus* (PO), *Lentinus squarrosulus* (LS), and a consortium of *Pleurotus ostreatus* and *Lentinus squarrosulus* (POLS) ranged from 2.09 – 9.99 %; 2.02 – 9.99 % and 2.31 – 9.99 % with the least recorded at 21, 28 and 28 days of degradation, respectively and their highest extractives were recorded before degradation. The highest hemicellulose contents of groundnut shell degraded by PO (30.39 %), LS (26.31 %), and POLS (26.07 %) were obtained on 14 days of degradation, and the least (15.09 %) was obtained when degraded by POLS for 35 days. The least lignin contents (33.43, 17.60, and 18.78 %) of all treatments (PO, LS, and POLS) were observed in the 49-day degraded sample, respectively. The cellulose content of groundnut shell degraded by PO, LS, and POLS ranged from 18.55 – 37.68 %, 23.44 – 52.61 %, and 21.00 – 52.69 %, respectively. The least and highest cellulose content by the degrading fungi were obtained at 14 and 49 days of degradation, respectively. Statistical analysis revealed that the extractives, hemicellulose, lignin, and cellulose contents of degraded groundnut shell by *Pleurotus ostreatus*, *Lentinus squarrosulus*, and a consortium of the two were significantly different ($P \leq 0.5$) with days of degradation. The component of degraded maize cob by all the treatments was in the order of hemicellulose > lignin > cellulose > extractives Table 2. The highest extractives of PO (8.78 %), LS (8.59 %), and POLS (10.33 %) were recorded in 7-day degraded maize cob. The highest hemicellulose contents of 40.70 %, 43.92 %, and 44.88 % were recorded when maize cob was degraded by PO, LS and POLS at 14, 63, and 14 days of degradation, respectively. The lignin content of maize cob degraded by PO, LS, and POLS ranged from 24.29 – 35.66 %, 26.45 – 35.37 %, and 22.65 – 35.85 %, respectively. The initial cellulose content (29.08 %) of undegraded maize cob was higher than those degraded except for maize cob degraded by PO for 49 days (29.13 %) which was not significantly different ($P > 0.05$) from the undegraded. The least cellulose content was observed at 14 days of degradation by PO (21.34 %) and POLS (15.07%). Maize cob degraded by LS had least cellulose content (16.22 %) at 49 days of degradation which was not significantly different ($P > 0.05$) from cellulose content (16.68 %) obtained at 14 days of degradation.

Table 1. Component of groundnut shell pretreated with *Pleurotus ostreatus* and *Lentinus squarrosulus* singly and combined.

Days	Extractives (%)			Hemicellulose (%)			Lignin (%)			Cellulose (%)		
	PO	LS	POLS	PO	LS	POLS	PO	LS	POLS	PO	LS	POLS
0	9.99^{e}	9.99^{d}	9.99^{d}	15.59^{a}	15.59^{a}	15.59^{a}	40.79^{c}	40.79^{c}	40.79^{c}	33.64^{f}	33.64^{g}	33.64^{f}
7	8.19^{d}	9.55^{d}	7.09^{bc}	28.41^{e}	26.18^{g}	24.44^{g}	43.06^{de}	35.38^{b}	37.13^{b}	20.34^{b}	28.90^{de}	31.35^{de}
14	3.01^{a}	2.90^{a}	3.13^{a}	30.39^{e}	26.31^{g}	26.07^{h}	48.05^{f}	47.35^{f}	49.80^{f}	18.55^{a}	23.44^{a}	21.00^{a}
21	2.09^{a}	3.47^{a}	3.41^{a}	21.40^{c}	20.91^{ef}	20.75^{de}	50.24^{g}	48.79^{f}	49.41^{f}	26.27^{c}	26.83^{bc}	26.43^{b}
28	2.45^{a}	2.02^{a}	2.31^{a}	16.81^{a}	16.81^{ab}	17.25^{b}	49.21^{fg}	51.57^{g}	50.20^{f}	31.52^{e}	29.59^{de}	30.24^{d}
35	5.74^{b}	6.31^{b}	6.31^{b}	18.88^{b}	18.89^{cd}	15.09^{a}	41.64^{cd}	43.27^{de}	41.53^{cd}	33.74^{f}	31.53^{f}	37.07^{g}
42	6.28^{bc}	7.62^{bc}	6.38^{b}	18.58^{b}	18.01^{bc}	19.60^{cd}	48.87^{fg}	48.50^{f}	47.84^{e}	26.27^{c}	25.87^{b}	26.18^{b}
49	7.95^{d}	7.58^{bc}	7.71^{bc}	20.94^{c}	22.21^{f}	20.81^{de}	33.43^{a}	17.60^{a}	18.78^{a}	37.68^{g}	52.61^{h}	52.69^{h}
56	7.68^{cd}	8.54^{cd}	8.23^{c}	23.92^{d}	21.02^{ef}	22.70^{f}	42.24^{cde}	42.33^{d}	40.94^{c}	26.16^{c}	28.10^{cd}	28.13^{c}
63	6.28^{bc}	7.14^{bc}	7.16^{bc}	24.43^{d}	19.84^{de}	18.34^{bc}	43.60^{e}	43.88^{e}	42.58^{d}	25.69^{c}	29.15^{de}	31.93^{e}
70	7.19^{bcd}	7.89^{c}	7.47^{bc}	24.86^{d}	21.19^{ef}	21.25^{e}	38.74^{b}	40.71^{c}	41.05^{c}	29.21^{d}	30.21^{ef}	30.24^{d}

Note: Mean values with different alphabetical superscripts along the column are significantly different ($P \le 0.05$).
PO: *Pleurotus ostreatus.*
LS: *Lentinus squarrosulus.*
POLS: Consortium of *Pleurotus ostreatus* and *Lentinus squarrosulus.*

Table 2. Component of maize cob pretreated with *Pleurotus ostreatus* and *Lentinus squarrosulus* singly and combined.

Days	Extractives (%)			Hemicellulose (%)			Lignin (%)			Cellulose (%)		
	PO	LS	POLS	PO	LS	POLS	PO	LS	POLS	PO	LS	POLS
0	8.26^{bc}	8.26^{c}	8.26^{c}	35.27^{ab}	35.27^{a}	35.27^{a}	27.38^{b}	27.38^{ab}	27.38^{b}	29.08^{e}	29.08^{f}	29.08^{e}
7	8.78^{c}	8.59^{c}	10.33^{d}	37.37^{cde}	42.80^{de}	39.73^{cd}	27.77^{b}	28.52^{b}	28.66^{bc}	26.08^{c}	20.09^{b}	21.28^{b}
14	3.59^{a}	5.10^{b}	4.20^{ab}	40.70^{g}	42.84^{de}	44.88^{f}	34.37^{de}	35.37^{e}	35.85^{h}	21.34^{a}	16.68^{a}	15.07^{a}
21	3.20^{a}	3.88^{b}	5.38^{b}	39.43^{fg}	40.02^{c}	39.54^{cd}	35.66^{e}	32.99^{d}	34.82^{gh}	21.72^{ab}	23.11^{d}	20.27^{b}
28	3.24^{a}	2.33^{a}	2.92^{a}	34.02^{a}	39.01^{c}	37.22^{b}	34.55^{de}	33.91^{de}	33.79^{fg}	28.20^{de}	24.74^{e}	26.07^{d}
35	8.62^{c}	7.39^{c}	7.40^{c}	36.39^{bc}	42.86^{de}	35.73^{a}	32.13^{c}	28.00^{b}	32.01^{e}	22.86^{b}	21.75^{cd}	24.86^{d}
42	6.93^{b}	8.12^{c}	7.34^{c}	36.71^{bc}	36.91^{b}	38.45^{bc}	33.37^{cd}	33.74^{d}	33.02^{ef}	22.99^{b}	21.22^{bc}	21.19^{b}
49	7.72^{bc}	7.64^{c}	7.79^{c}	38.86^{ef}	41.75^{d}	41.49^{e}	24.29^{a}	34.39^{de}	29.39^{cd}	29.13^{e}	16.22^{a}	21.33^{b}
56	7.90^{bc}	8.55^{c}	8.60^{c}	38.54^{def}	38.91^{c}	42.05^{e}	26.72^{b}	31.23^{c}	28.29^{bc}	26.83^{cd}	21.31^{bc}	21.07^{b}
63	6.95^{b}	7.61^{c}	7.61^{c}	38.22^{def}	43.92^{e}	38.98^{c}	26.68^{b}	26.45^{a}	30.57^{d}	28.15^{de}	22.02^{cd}	22.84^{c}
70	7.03^{b}	7.56^{c}	7.87^{c}	37.22^{cd}	36.91^{b}	40.91^{de}	27.29^{b}	27.60^{ab}	22.65^{a}	28.46^{e}	27.93^{f}	28.57^{e}

Note: Mean values with different alphabetical superscripts along the column are significantly different ($P \le 0.05$)
PO: *Pleurotus ostreatus.*
LS: *Lentinus squarrosulus.*
POLS: Consortium of *Pleurotus ostreatus* and *Lentinus squarrosulus.*

Table 3. Component of maize straw pretreated with *Pleurotus ostreatus* and *Lentinus squarrosulus* singly and combined.

Days	Extractives (%)			Hemicellulose (%)			Lignin (%)			Cellulose (%)		
	PO	LS	POLS	PO	LS	POLS	PO	LS	POLS	PO	LS	POLS
0	10.72^{g}	10.72^{f}	10.72^{e}	42.05^{a}	42.05^{ab}	42.05^{def}	29.52^{e}	29.52^{b}	29.52^{c}	17.71^{g}	17.71^{ef}	17.71^{a}
7	2.87^{b}	3.18^{a}	4.98^{a}	48.29^{cd}	42.73^{bc}	39.35^{c}	26.18^{b}	30.53^{b}	18.42^{a}	22.66^{h}	23.56^{g}	37.24^{e}
14	4.59^{c}	5.22^{b}	5.30^{ab}	47.55^{bc}	46.47^{f}	36.14^{b}	31.00^{f}	35.99^{d}	33.44^{d}	16.86^{fg}	12.33^{ab}	25.12^{d}
21	7.39^{de}	6.66^{bc}	6.40^{abc}	46.34^{b}	43.41^{bcd}	40.77^{d}	32.81^{g}	38.99^{f}	35.23^{e}	13.46^{d}	10.94^{a}	17.60^{a}
28	1.12^{a}	2.00^{a}	13.97^{f}	49.78^{e}	45.11^{ef}	33.40^{a}	37.42^{h}	37.59^{e}	35.13^{e}	11.68^{c}	15.30^{d}	17.50^{a}
35	9.05^{f}	8.67^{e}	8.05^{d}	46.57^{b}	41.14^{a}	43.77^{g}	29.12^{de}	27.14^{a}	25.70^{b}	15.27^{e}	23.05^{g}	22.48^{c}
42	7.06^{de}	6.93^{cd}	6.65^{bcd}	50.66^{ef}	43.55^{cd}	41.40^{de}	36.69^{h}	36.06^{d}	35.76^{e}	5.60^{a}	13.46^{bc}	16.19^{a}
49	7.65^{ef}	8.34^{de}	7.00^{cd}	51.62^{f}	43.70^{cde}	42.78^{efg}	30.98^{f}	33.42^{c}	33.68^{d}	9.75^{b}	14.55^{cd}	16.54^{a}
56	7.62^{ef}	7.32^{cde}	7.47^{cd}	49.5^{de}	44.34^{de}	42.97^{fg}	27.77^{cd}	29.51^{b}	29.73^{c}	15.12^{e}	18.84^{f}	19.83^{b}
63	6.03^{d}	6.30^{bc}	6.62^{bcd}	53.33^{g}	46.35^{f}	43.54^{fg}	27.05^{bc}	30.65^{b}	26.41^{b}	13.59^{d}	16.70^{e}	23.43^{c}
70	8.29^{ef}	7.01^{cd}	6.94^{cd}	54.16^{g}	43.59^{cd}	42.75^{efg}	21.84^{a}	30.97^{b}	30.16^{c}	15.71^{ef}	18.43^{f}	20.15^{b}

Note: Mean values with different alphabetical superscripts along the column are significantly different ($P \leq 0.05$)
PO: *Pleurotus ostreatus.*
LS: *Lentinus squarrosulus.*
POLS: Consortium of *Pleurotus ostreatus* and *Lentinus squarrosulus.*

Table 4. Component of sugarcane bagasse pretreated with *Pleurotus ostreatus* and *Lentinus squarrosulus* singly and combined.

Days	Extractives (%)			Hemicellulose (%)			Lignin (%)			Cellulose (%)		
	PO	LS	POLS	PO	LS	POLS	PO	LS	POLS	PO	LS	POLS
0	11.12^{f}	11.12^{e}	11.12^{g}	36.15^{a}	36.15^{a}	36.15^{a}	29.71^{b}	29.71^{c}	29.71^{e}	23.01^{def}	23.01^{c}	23.01^{d}
7	4.45^{ab}	3.52^{ab}	4.48^{b}	35.44^{a}	38.33^{b}	39.34^{b}	27.27^{a}	27.49^{b}	26.10^{bc}	32.85^{h}	30.66^{f}	30.08^{g}
14	3.67^{ab}	2.35^{a}	2.43^{a}	35.21^{a}	39.46^{bc}	42.17^{cd}	33.35^{c}	34.70^{e}	35.77^{g}	27.78^{g}	23.48^{c}	19.62^{c}
21	4.83^{bc}	4.39^{b}	5.38^{bc}	40.51^{c}	40.36^{cd}	45.03^{f}	32.86^{c}	30.00^{c}	34.63^{g}	21.80^{cd}	25.25^{d}	14.95^{a}
28	3.01^{a}	3.27^{ab}	2.19^{a}	39.02^{b}	42.25^{e}	41.12^{c}	33.64^{c}	32.10^{d}	32.46^{f}	24.33^{f}	22.38^{c}	24.23^{de}
35	8.73^{e}	7.17^{cd}	4.80^{b}	41.84^{cd}	39.67^{bcd}	43.66^{ef}	25.88^{a}	29.88^{c}	27.91^{d}	23.55^{ef}	23.29^{c}	23.63^{de}
42	6.72^{d}	7.08^{cd}	6.31^{cd}	40.63^{c}	40.50^{cd}	41.55^{c}	34.23^{c}	32.21^{d}	33.07^{f}	18.41^{b}	20.21^{b}	19.07^{c}
49	9.26^{e}	7.96^{d}	7.81^{ef}	40.56^{c}	41.21^{de}	43.84^{ef}	33.35^{c}	33.01^{d}	31.62^{f}	16.83^{a}	17.83^{a}	16.73^{b}
56	9.98^{ef}	11.17^{e}	8.92^{f}	42.57^{de}	40.98^{cde}	41.43^{c}	26.88^{a}	26.89^{b}	24.95^{b}	20.57^{c}	20.97^{b}	24.70^{e}
63	6.00^{cd}	6.00^{c}	7.40^{de}	42.52^{de}	44.17^{f}	41.32^{c}	29.01^{b}	26.41^{b}	27.16^{cd}	22.47^{de}	23.42^{c}	24.12^{de}
70	6.77^{d}	6.87^{cd}	7.35^{de}	43.66^{e}	40.69^{cd}	43.06^{de}	26.98^{a}	23.91^{a}	23.21^{a}	22.58^{de}	28.53^{e}	26.38^{g}

Note: Mean values with different alphabetical superscripts along the column are significantly different ($P \leq 0.05$)
PO: *Pleurotus ostreatus.*
LS: *Lentinus squarrosulus.*
POLS: Consortium of *Pleurotus ostreatus* and *Lentinus squarrosulus.*

The Extractives, hemicellulose, lignin, and cellulose contents of degraded maize straw is as shown in Table 3. The highest extractive (10.72 %) was recorded in undegraded maize straw and the lowest extractives of 1.12 %, 2.00 %, and 4.98 % were observed in PO, LS, and POLS-degraded maize straw at 28, 28, and 7 days of degradation, respectively. Hemicellulose content ranged from 42.05 – 54.16 %, 41.14 – 46.47 %, and 33.40 – 43.77 % in PO, LS, and POLS degraded maize straw, respectively. The hemicellulose content of PO-degraded maize straw was higher than LS and POLS-degraded maize straw throughout the period of degradation. The observed highest lignin contents by maize straw degraded by PO (37.42 %), LS (38.99 %), and POLS (35.76%) were recorded at 28, 21, and 42 days of degradation, respectively. It was observed that maize straw degraded for 7 days had highest cellulose content for PO (22.66 %), LS (23.56 %), and POLS (37.24 %). Days of degradation had a significant effect ($P \leq 0.05$) on extractives, hemicellulose, lignin, and cellulose of degraded maize straw.

The component of pretreated sugarcane bagasse is shown in Table 4. From the table, undegraded sugarcane bagasse had the highest extractives when compared with those degraded by selected white rot fungi. The least extractive observed in sugarcane bagasse degraded by PO, LS, and POLS were 3.01 %, 2.35 %, and 2.19 % at 28, 14, and 28 days of degradation, respectively. Hemicellulose contents of 35.21 – 43.66 %, 36.15 – 44.17 %, and 36.15 – 45.03 % were observed in PO, LS, and POLS-degraded sugarcane bagasse with their highest content at 70, 63, and 21 days of degradation, respectively. While the highest lignin content of PO-degraded maize straw (34.23 %) was observed at 42 days of degradation, the highest lignin contents of LS (34.70 %) and POLS (35.77%) -degraded sugarcane bagasse were recorded at 14 days of degradation. The least cellulose content obtained when sugar cane bagasse was degraded by PO, LS, and POLS were 16.83 %, 17.83 %, and 14.95 % at 49, 49, and 21 days of degradation, respectively. There were significant differences ($P \leq 0.05$) in the values of extractives, hemicellulose, lignin, and cellulose with the day of degradation.

Table 5 shows the extractives, hemicellulose, lignin, and cellulose contents of PO, LS, and POLS-degraded rice straw. The extractives of rice straw degraded by PO, LS, and POLS ranged from 3.20 – 8.86 %, 3.84 – 8.86 %, and 3.07 – 8.86 %, respectively. Hemicellulose content of 39.61 – 56.70 %, 39.61 – 50.38 %, and 36.95 – 51.66 % were recorded in PO, LS, and POLS-degraded rice straw, respectively. During degradation, ranges of 20.91 – 31.54 %, 24.91 – 34.85 %, and 24.37 – 35.19 % of lignin content were obtained in PO, LS, and POLS-degraded rice straw, respectively. The cellulose content of rice straw degraded by PO ranged from 12.16 to 27.97 % with the least and highest at 70 and 7 days of degradation, respectively. Cellulose content range of 8.06 – 25.25 % and 15.31 – 30.46 % were recorded in LS and POLS degraded rice straw, respectively. Statistical analysis revealed values of extractives, hemicellulose, lignin, and cellulose of degraded rice are significantly different ($P \leq 0.05$) with days of degradation.

The reducing sugar content of lignocellulosic samples (groundnut shell, maize cob, maize straw, sugar cane bagasse, and rice straw) determined every 7 days during 70 days of degradation is shown in Table 6. Reducing sugar of PO-pretreated groundnut shell ranged from 2.61 to 11.23 mg/g with the least and highest at 56 and 49 days of degradation, respectively. The least reducing sugar (0.78 mg/g) of groundnut shell degraded by LS was obtained at 70 days of degradation and the highest (11.83 mg/g) at 21 days of degradation. The reducing sugar content of POLS-degraded groundnut shell ranged from 2.90 to 8.01 mg/g with the least and highest at 7 and 49 days of degradation, respectively, and were not significantly different ($P > 0.05$). Reducing sugar of maize cob degraded by PO, LS, and POLS ranged from 0.37 – 8.16 mg/g, 3.64 – 13.32 mg/g, and 3.76 – 13.25 mg/g, respectively. The highest reducing sugar contents of PO, LS, and POLS-degraded maize straw were 27.03 mg/g, 20.41 mg/g, and 19.70 mg/g at 14, 35, and 0 days, respectively. The reducing sugar content of PO-degraded maize straw was generally higher than the reducing sugar of LS, and POLS-degraded maize straw. Sugarcane bagasse degraded by PO, LS, and POLS have reducing sugar in the range of 9.08 – 28.70 mg/g, 11.75 – 25.58 mg/g, and 10.28 – 31.94 mg/g with their highest content at 7, 7, and 49 days, respectively. The lowest reducing sugar (6.88 mg/g) was recorded in rice straw before degradation.

Table 5. Component of rice straw pretreated with *Pleurotus ostreatus* and *Lentinus squarrosulus* singly and combined.

Days	Extractives (%)			Hemicellulose (%)			Lignin (%)			Cellulose (%)		
	PO	LS	POLS	PO	LS	POLS	PO	LS	POLS	PO	LS	POLS
0	8.86^{d}	8.86^{d}	8.86^{c}	39.61^{a}	39.61^{a}	39.61^{b}	26.95^{f}	26.95^{c}	26.95^{b}	24.59^{f}	24.59^{fg}	24.59^{d}
7	4.26^{a}	3.84^{a}	3.58^{a}	46.87^{c}	39.97^{a}	36.95^{a}	20.91^{a}	30.99^{d}	29.01^{c}	27.97^{g}	25.20^{g}	30.46^{e}
14	3.31^{a}	6.71^{bc}	3.40^{a}	51.37^{d}	50.38^{f}	43.23^{cd}	31.54^{h}	34.85^{e}	35.19^{f}	13.79^{b}	8.06^{a}	18.18^{b}
21	6.53^{b}	4.86^{a}	4.46^{a}	45.29^{b}	48.96^{ef}	48.18^{f}	25.61^{ef}	31.54^{d}	31.19^{d}	22.57^{e}	14.64^{c}	16.17^{a}
28	3.20^{a}	4.53^{a}	3.07^{a}	52.40^{de}	49.44^{ef}	45.91^{e}	30.16^{gh}	34.79^{e}	32.93^{e}	14.24^{b}	11.24^{b}	18.09^{b}
35	8.19^{cd}	7.85^{bcd}	7.60^{bc}	51.37^{d}	48.52^{e}	48.29^{f}	23.68^{cd}	26.72^{bc}	28.79^{c}	16.76^{c}	16.91^{d}	15.31^{a}
42	8.21^{cd}	6.94^{bc}	7.97^{bc}	45.36^{b}	45.33^{d}	44.17^{d}	29.41^{g}	34.13^{e}	32.17^{de}	17.03^{c}	13.61^{c}	15.70^{a}
49	7.16^{bc}	6.50^{b}	7.22^{b}	47.86^{c}	41.41^{b}	42.08^{c}	24.88^{de}	30.27^{d}	30.75^{d}	20.11^{d}	21.82^{e}	19.95^{c}
56	8.17^{cd}	8.18^{cd}	7.77^{bc}	53.66^{e}	42.80^{bc}	46.18^{e}	24.96^{de}	27.17^{c}	26.90^{b}	13.21^{ab}	21.85^{e}	19.15^{bc}
63	7.98^{bcd}	6.73^{bc}	8.34^{bc}	53.65^{e}	42.62^{b}	48.77^{f}	22.08^{ab}	25.40^{ab}	26.23^{b}	16.29^{c}	25.25^{g}	16.66^{a}
70	7.97^{bcd}	7.21^{bc}	8.11^{bc}	56.70^{f}	44.14^{cd}	51.66^{g}	23.17^{bc}	24.91^{a}	24.37^{a}	12.16^{a}	23.73^{f}	15.86^{a}

Note: Mean values with different alphabetical superscripts along the column are significantly different ($P \leq 0.05$).
PO: *Pleurotus ostreatus.*
LS: *Lentinus squarrosulus.*
POLS: Consortium of *Pleurotus ostreatus* and *Lentinus squarrosulus.*

Table 6. Reducing sugar content (mg/g) of lignocellulosic substrates degraded by *Lentinus squarrosulus* and *Pleurotus ostreatus* singly and combined.

Period (Days)	GS			MC			MS			SB			RS		
	PO	LS	POLS	PO	LS	POLS	PO	LS	POLS	PO	LS	POLS	PO	LS	POLS
0	3.99^{ab}	3.99^{ab}	3.99^{a}	3.76^{abcd}	3.76^{a}	3.76^{a}	19.70^{a}	19.70^{b}	19.70^{b}	16.88^{abc}	16.88^{abc}	16.88^{ab}	6.88^{a}	6.88^{a}	6.88^{a}
7	6.37^{abc}	4.54^{ab}	2.90^{a}	7.06^{cd}	13.26^{b}	10.67^{a}	14.76^{a}	11.64^{ab}	12.39^{ab}	28.70^{d}	25.58^{c}	23.68^{abc}	17.80^{bc}	15.68^{a}	15.74^{ab}
14	4.20^{abc}	5.16^{ab}	3.12^{a}	3.32^{abcd}	8.45^{ab}	6.68^{a}	27.03^{a}	15.16^{ab}	9.14^{a}	22.66^{cd}	14.53^{ab}	10.28^{a}	19.04^{bcd}	11.19^{a}	15.80^{ab}
21	7.14^{bc}	11.83^{c}	6.36^{a}	4.58^{abcd}	6.49^{ab}	8.59^{a}	22.26^{a}	18.81^{ab}	13.29^{ab}	16.23^{abc}	22.66^{bc}	23.13^{abc}	31.84^{ef}	11.57^{a}	20.70^{ab}
28	3.42^{ab}	8.56^{bc}	7.88^{a}	4.45^{abcd}	3.85^{a}	5.82^{a}	18.24^{a}	15.91^{ab}	14.12^{ab}	11.52^{ab}	22.17^{bc}	12.58^{ab}	25.88^{cde}	10.27^{a}	15.17^{ab}
35	8.08^{cd}	3.44^{a}	7.03^{a}	4.74^{abcd}	9.34^{ab}	9.66^{a}	24.94^{a}	20.41^{b}	18.93^{b}	19.51^{bc}	18.90^{abc}	17.68^{abc}	30.44^{ef}	13.46^{a}	19.27^{ab}
42	5.67^{abc}	1.71^{a}	4.98^{a}	0.37^{a}	13.32^{b}	10.73^{a}	18.14^{a}	16.29^{ab}	14.27^{ab}	9.61^{a}	17.59^{abc}	13.03^{ab}	11.93^{ab}	17.01^{a}	17.92^{ab}
49	11.23^{d}	4.08^{ab}	8.01^{a}	5.76^{bcd}	7.37^{ab}	8.92^{a}	11.93^{a}	16.76^{ab}	11.80^{ab}	16.00^{abc}	19.88^{abc}	31.94^{c}	37.96^{f}	12.55^{a}	11.38^{ab}
56	2.61^{a}	5.64^{ab}	7.99^{a}	2.89^{abc}	4.92^{ab}	5.20^{a}	15.54^{a}	16.70^{ab}	11.86^{ab}	11.08^{ab}	20.26^{abc}	21.73^{abc}	19.85^{bcd}	14.00^{a}	17.45^{ab}
63	3.55^{ab}	1.51^{a}	7.26^{a}	8.16^{d}	7.62^{ab}	13.25^{a}	17.48^{a}	13.72^{ab}	11.25^{ab}	18.60^{abc}	11.75^{a}	26.37^{bc}	27.47^{de}	13.37^{a}	28.74^{b}
70	5.50^{abc}	0.78^{a}	6.25^{a}	0.76^{ab}	3.64^{a}	7.12^{a}	16.96^{a}	9.66^{a}	12.58^{ab}	9.08^{a}	15.01^{ab}	20.80^{abc}	18.79^{bcd}	8.34^{a}	18.14^{ab}

Note: Mean values with different alphabetical superscripts along the column are significantly different ($P \leq 0.05$).
GS: Groundnut shell.
MC: Maize cob.
MS: Maize straw.
SB: Sugarcane bagasse.
RS: Rice straw.
PO: *Pleurotus ostreatus.*
LS: *Lentinus squarrosulus.*
POLS: Consortium of *Pleurotus ostreatus* and *Lentinus squarrosulus.*

The highest reducing sugar of rice straw degraded by PO (37.96 mg/g), LS (17.01 mg/g), and POLS (28.74 mg/g) were recorded at 49, 42, and 63 days, respectively. Statistical analysis revealed that there was no significant difference ($P>0.05$) in the reducing sugar content of rice straw that was degraded by Lentinus squarrosulus with days of degradation.

4. DISCUSSIONS

Changes in chemical composition observed when different lignocellulolytic substrates were degraded with *Pleurotus ostreatus* and *Lentinus squarrosulus* through solid-state fermentation could be due to the metabolites (cellulase, xylanase, lignase/laccase, etc.) produced by these organisms, which can degrade different parts of lignocellulose. This observation corroborates the work of Issaka, et al. [17] and Wuanor and Ayoade [18] who degraded groundnut shell with *Pleurotus* species and reported changes in the chemical composition of groundnut shell. Costa-Silva, et al. [19] also observed changes in the composition of grape stalks degraded by some white rot fungi. Lower extractives recorded in most of the degraded substrates than in non-degraded ones might probably be due to the utilization of the extractives as nutrients during degradation by these mushrooms [20]. The values of extractives vary from biomass to biomass and between different parts of the same plant [20, 21].

Higher hemicellulose content observed in degraded lignocellulolytic substrates compared with non-degraded might be a result of low required nutrients needed for the production of hemicellulases (xylanase and others) on the substrates that could have converted hemicellulose to glucose and xylose. This is contrary to the findings of Issaka, et al. [17] and Wuanor and Ayoade [18] who recorded a decrease in hemicellulose content after degrading groundnut shell with *Pleurotus* species. The percentage composition of lignocellulolytic substrates differs from one another based on the class of the substrate which is softwood or hardwood [20]. Generally, lower hemicellulose content was observed when degraded by co-culture of *Pleurotus ostreatus* and *Lentinus squarrosulus* than when degraded singly. This might be due to the synergistic relationship between *Pleurotus ostreatus* and *Lentinus squarrosulus* in the utilization of hemicellulose. There have been reports that organisms performed differently when in consortium from when used singly [22]. The decrease in lignin content of groundnut shell observed after 49 days of degradation by *Pleurotus ostreatus*, *Lentinus squarrosulus* and consortium of *Pleurotus ostreatus* and *Lentinus squarrosulus* showed that these mushrooms can remove lignin bonds that prevent holocellulose from being broken down to simple and fermentable sugar. This observation has been reported to be due to the production of lignin-degrading enzymes by these organisms [23-26]. A similar observation of a decrease in lignin content of degraded groundnut shell by *Pleurotus ostreatus* for 5 weeks [17] and 30 days [18] has also been reported.

Conversion of cellulose to simple sugars by cellulase-producing mushrooms selected for degradation in this work could be responsible for a decrease in cellulose content observed at most sampling times in all selected lignocellulolytic substrates. The cellulose part of lignocellulosic substrates would have been extensively utilized and converted to hexoses by selected mushrooms leading to a decrease in cellulose after degradation as reported by some researchers [27]. A similar observation of a decrease in cellulose content after degradation with *Pleurotus* species was reported by Akinfemi [28] and Huang, et al. [29] from maize cob and crop straw respectively.

The higher reducing sugar released from groundnut shell degraded by the monoculture of *Pleurotus ostreatus* and *Lentinus squarrosulus* than co-culture of the two might be due to the high utilization of the released reducing sugar as carbon and energy sources by the co-culture than monoculture [30, 31] or the organisms might be having an antagonistic effect on each other leading to decrease in released reducing sugar when grown together. While the observed higher reducing sugar released in *Pleurotus ostreatus* and *Lentinus squarrosulus*-degraded maize cob than non-degraded one was probably because of the interaction between hydrolytic and oxidative enzymes released by these organisms when degrading maize cob, breaking down cellulose and hemicellulose to simple sugar [32, 33]. A similar observation of increased reducing sugar content of maize cob, when degraded by *Pleurotus ostreatus*, was reported by Adamafio, et al. [32]. The increase in reducing sugar content observed in degraded maize straw could be due to the breaking down of different components of maize straw to reducing sugar by the enzymes produced by the organisms which could be influenced by both genetic makeup and environmental conditions [29, 34, 35]. Higher reducing sugar content observed in degraded sugarcane bagasse could be due to the ability of *Pleurotus ostreatus* and *Lentinus squarrosulus* to produce cellulase and xylanase which could have broken the holocellulose content of sugarcane bagasse to reducing sugar Ravichandran, et al. [26]; Jonathan and Akinfemi [36]; Dong, et al. [37]; Shankarappa, et al. [38]; Gani, et al. [39]. Gani, et al. [39] reported high reducing sugar when sugarcane bagasse was pretreated with alkaline and acid. The ability of *Pleurotus ostreatus* and *Lentinus squarrosulus* to produce lignocellulolytic enzymes that can breakdown cellulose, hemicellulose, and lignin to simple sugar could be responsible for high amount of reducing sugar recorded in degraded rice straw Jonathan and Akinfemi [36]; Belal [40]; Nurika, et al. [41]. Belal [40] reported high reducing sugar in rice straw degraded with *Trichoderma reesei* for 14 days while Nurika, et al. [41] observed a higher amount of reducing sugar after 21 days of degradation of rice straw with *Serpula lacrymans*.

5. CONCLUSION

The ability of *Pleurotus ostreatus* and *Lentinus squarrosulus* to degrade lignocellulosic substrates to simple sugars shows that these organisms could be employed in second-generation biofuel production where simple sugars released from lignocellulose would be used for ethanol production. Highest reducing sugar content (37.96 mg/g) was obtained by degrading rice straw by *Pleurotus ostreatus* for 49 days. Sole degradation with *Pleurotus ostreatus* had a better yield of reducing sugar than *Lentinus squarrosulus* and co-cultured. The amount of reducing sugar released varied with substrates, organisms, and degradation time.

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REFERENCES

[1] S. M. Wakil, O. J. Aladekoyi, S. A. Fasiku, and C. O. Adenipekun, "Production of bioethanol from lignocellulosic waste," *Nigerian Journal of Science*, vol. 51, pp. 23-35, 2017.

[2] S. A. Fasiku and S. Monilola Wakil, "Pretreatment of maize straw with Pleurotus ostreatus and Lentinus squarrosulus for bioethanol production using Saccharomyces cerevisiae," *Novel Research in Microbiology Journal*, vol. 5, no. 6, pp. 1480-1493, 2021. https://doi.org/10.21608/nrmj.2021.209731

[3] S. A. Fasiku and S. M. Wakil, "Screening of factors responsible for conversion of maize straw into bioethanol," *Journal of Microbiology, Biotechnology and Food Sciences*, vol. 12, no. 2, pp. e5901-e5901, 2022. https://doi.org/10.55251/jmbfs.5901

[4] A. Kumar and R. Chandra, "Ligninolytic enzymes and its mechanisms for degradation of lignocellulosic waste in environment," *Heliyon*, vol. 6, no. 2, p. e03170, 2020. https://doi.org/10.55251/jmbfs.5901

[5] C. A. Cardona and Ó. J. Sánchez, "Fuel ethanol production: process design trends and integration opportunities," *Bioresource Technology*, vol. 98, no. 12, pp. 2415-2457, 2007.

[6] V. B. Agbor, N. Cicek, R. Sparling, A. Berlin, and D. B. Levin, "Biomass pretreatment: Fundamentals toward application," *Biotechnology Advances*, vol. 29, no. 6, pp. 675-685, 2011.

[7] S. Rezania *et al.*, "Different pretreatment technologies of lignocellulosic biomass for bioethanol production: An overview," *Energy*, vol. 199, p. 117457, 2020. https://doi.org/10.1016/j.energy.2020.117457

[8] V. K. Nguyen *et al.*, "Review on pretreatment techniques to improve anaerobic digestion of sewage sludge," *Fuel*, vol. 285, p. 119105, 2021.

[9] A. K. Kumar and S. Sharma, "Recent updates on different methods of pretreatment of lignocellulosic feedstocks: A review," *Bioresources and Bioprocessing*, vol. 4, no. 1, pp. 1-19, 2017. https://doi.org/10.1186/s40643-017-0137-9

[10] R. Millati, S. Syamsiah, C. Niklasson, M. N. Cahyanto, K. Ludquist, and M. J. Taherzadeh, "Biological pretreatment of lignocelluloses with white-rot fungi and its applications: A review," *BioResources*, vol. 6, no. 4, pp. 5224-5259, 2011. https://doi.org/10.15376/biores.6.4.isroi

[11] A. Hatakka and K. Hammel, "Fungal biodegradation of lignocelluloses," *Mycota-A Comprehensive Treatise on Fungi as Experimental Systems for Basic and Applied Research*, vol. 10, pp. 319-340, 2010. https://doi.org/10.1007/978-3-642-11458-8_15

[12] H. Suryadi *et al.*, "Biodelignification of lignocellulose using ligninolytic enzymes from white-rot fungi," *Heliyon*, vol. 8, no. 2, p. e08865, 2022. https://doi.org/10.1016/j.heliyon.2022.e08865

[13] M. Ganash, T. M. A. Ghany, M. A. Al Abboud, M. M. Alawlaqi, H. Qanash, and B. H. Amin, "Lignocellulolytic activity of Pleurotus ostreatus under solid state fermentation using silage, stover, and cobs of maize," *BioResources*, vol. 16, no. 2, pp. 3797-3807, 2021. https://doi.org/10.15376/biores.16.2.3797-3807

[14] M. Marinovíc *et al.*, "Comparative analysis of enzyme production patterns of lignocellulose degradation of two white rot fungi: Obba rivulosa and Gelatoporia subvermispora," *Biomolecules*, vol. 12, no. 8, p. 1017, 2022. https://doi.org/10.3390/biom12081017

[15] L. Lin, R. Yan, Y. Liu, and W. Jiang, "In-depth investigation of enzymatic hydrolysis of biomass wastes based on three major components: Cellulose, hemicellulose and lignin," *Bioresource Technology*, vol. 101, no. 21, pp. 8217-8223, 2010. https://doi.org/10.1016/j.biortech.2010.05.084

[16] G. L. Miller, "Use of dinitrosalicylic acid reagent for determination of reducing sugar," *Analytical Chemistry*, vol. 31, no. 3, pp. 426-428, 1959. https://doi.org/10.1021/ac60147a030

[17] J. Issaka, F. Alemawor, and V. P. Dzogbefia, "Bioconversion impact of Pleurotus ostreatus on the value of rice and groundnut by-products as feed resources," *Research in Biotechnology*, vol. 4, no. 5, pp. 24-30, 2013. https://doi.org/10.15193/zntj/2013/90/119-128

[18] A. Wuanor and J. Ayoade, "Performance of West African dwarf goats fed Pleurotus tuber-regium biodegraded rice straw and maize offal: Brewer yeast slurry mixture," *Journal of Experimental Agriculture International*, vol. 17, no. 1, pp. 1-10, 2017. https://doi.org/10.9734/jeai/2017/34398

[19] V. Costa-Silva *et al.*, "Biovalorization of grape stalks as animal feed by solid state fermentation using white-rot fungi," *Applied Sciences*, vol. 12, no. 13, p. 6800, 2022. https://doi.org/10.3390/app12136800

[20] M. B. Pecha and M. Garcia-Perez, "Chapter 29 - pyrolysis of lignocellulosic biomass: Oil, char, and gas, Editor(s): Anju Dahiya, bioenergy," 2nd ed.: Academic Press, 2020, pp. 581-619. https://doi.org/10.1016/B978-0-12-815497-7.00029-4

[21] P. Bajpai, "Chapter 2 - wood and fiber fundamentals, Editor(s): Pratima Bajpai, Biermann's handbook of pulp and paper," 3rd ed.: Elsevier, 2018, pp. 19-74. https://doi.org/10.1016/B978-0-12-814240-0.00002-1

[22] W. Wang, T. Yuan, and B. Cui, "Biological pretreatment with white rot fungi and their co-culture to overcome lignocellulosic recalcitrance for improved enzymatic digestion," *BioResources*, vol. 9, no. 3, pp. 3968-3976, 2014. https://doi.org/10.15376/biores.9.3.3968-3976

[23] O. S. Isikhuemhen, N. A. Mikiashvili, C. O. Adenipekun, E. I. Ohimain, and G. Shahbazi, "The tropical white rot fungus, Lentinus squarrosulus Mont: lignocellulolytic enzymes activities and sugar release from cornstalks under solid state fermentation," *World Journal of Microbiology and Biotechnology*, vol. 28, pp. 1961-1966, 2012. https://doi.org/10.1007/s11274-011-0998-6

[24] R. Radhika, G. R. Jebapriya, and J. J. Gnanadoss, "Production of cellulase and laccase using Pleurotus sp. under submerged and solid-state fermentation," *International Journal of Current Science*, vol. 6, pp. 7-13, 2013. https://doi.org/10.1002/elsc.200700039

[25] X. Liu, W. Deng, and Y. Yang, "Characterization of a novel laccase LAC-Yang1 from white-rot fungus Pleurotus ostreatus strain Yang1 with a strong ability to degrade and detoxify chlorophenols," *Molecules*, vol. 26, no. 2, p. 473, 2021. https://doi.org/10.3390/molecules26020473

[26] A. Ravichandran, R. Rao, S. Gopinath, and M. Sridhar, "Augmenting versatile peroxidase production from lentinus squarrosulus and its role in enhancing ruminant feed," *BioResources*, vol. 16, pp. 1600-1615, 2021. https://doi.org/10.15376/biores.16.1.1600-1615

[27] Y. Metri, L. Warly, and Syitman, "Biodegradation of lignin by white rot fungi (Pleurotus ostreatus) to decrease the fibre components in the palm midrib," *Pakistan Journal of Nutrition*, vol. 17, no. 2, pp. 71-75, 2018. https://doi.org/10.3923/pjn.2018.71.75

[28] A. Akinfemi, "Nutritive value and in vitro gas production of fungal treated maize cobs," *African Journal of Food, Agriculture, Nutrition and Development*, vol. 10, no. 8, pp. 2943-2955, 2010. https://doi.org/10.4314/ajfand.v10i8.60878

[29] L. Huang, N. Sun, L. Ban, Y. Wang, and H. Yang, "Ability of different edible fungi to degrade crop straw," *AMB Express*, vol. 9, no. 1, pp. 1-6, 2019. https://doi.org/10.1186/s13568-018-0731-z

[30] O. B. Akpor, "Dye decolouration by immobilized and free bacterial cells at different glucose concentration," *Research Journal of Environmental Sciences*, vol. 12, no. 1, pp. 33-40, 2018. https://doi.org/10.3923/rjes.2018.33.40

[31] H. Hu *et al.*, "Glucose monitoring in living cells with single fluorescent protein-based sensors," *RSC Advances*, vol. 8, no. 5, pp. 2485-2489, 2018. https://doi.org/10.1039/c7ra11347a

[32] N. A. Adamafio, M. Obodai, and B. Brimpong, "Solid state fermentation of maize (Zea mays) cob by Pleurotus ostreatus strain EM-1: Biopolymer profiles and cellulose degradability," *International Journal of Biological and Chemical Sciences*, vol. 3, no. 6, pp. 1459-1466, 2009. https://doi.org/10.4314/ijbcs.v3i6.53169

[33] O. A. Ogunyewo and F. M. Olajuyigbe, "Unravelling the interactions between hydrolytic and oxidative enzymes in degradation of lignocellulosic biomass by Sporothrix carnis under various fermentation conditions," *Biochemistry Research International*, vol. 2016, p. 1614370, 2016. https://doi.org/10.1155/2016/1614370

[34] W. Huang *et al.*, "Effect of physicochemical pretreatments and enzymatic hydrolysis on corn straw degradation and reducing sugar yield," *BioResources*, vol. 12, no. 4, pp. 7002-7015, 2017. https://doi.org/10.15376/biores.12.4.7002-7015

[35] H. Wu, T. Nakazawa, R. Morimoto, M. Sakamoto, and Y. Honda, "Targeted disruption of hir1 alters the transcriptional expression pattern of putative lignocellulolytic genes in the white-rot fungus Pleurotus ostreatus," *Fungal Genetics and Biology*, vol. 147, p. 103507, 2021. https://doi.org/10.1016/j.fgb.2020.103507

[36] S. G. Jonathan and A. Akinfemi, "Chemical compositions of Zea mays cobs biodegraded by Lentinus subnudus and Pleurotus tuber-regium," *BioTechnology*, vol. 5, pp. 60-65, 2010.

[37] X. Q. Dong, J. S. Yang, N. Zhu, E. T. Wang, and H. L. Yuan, "Sugarcane bagasse degradation and characterization of three white-rot fungi," *Bioresource Technology*, vol. 131, pp. 443-451, 2013. https://doi.org/10.1016/j.biortech.2012.12.182

[38] T. H. Shankarappa, G. S. Geeta, M. J. Manju, C. R. Patil, H. Vamadevaiah, and K. S. Jagadeesh, "Saccharification of alkali pretreated agroresidues to fermentable sugars by crude enzymes of cellulolytic fungi," *Asian Journal of Biological and Life Sciences*, vol. 4, no. 2, pp. 114-121, 2015.

[39] M. Gani, N. Abdulkadir, S. B. Usman, H. M. Maiturare, and S. Gabriel, "Production of bioethanol from sugarcane bagasse using Saccharomyces cerevisiae," *Biotechnology Journal International*, vol. 22, pp. 1-8, 2018.

[40] E. B. Belal, "Bioethanol production from rice straw residues," *Brazilian Journal of Microbiology*, vol. 44, no. 1, pp. 225-234, 2013. https://doi.org/10.1590/s1517-83822013000100033

[41] I. Nurika, S. Suhartini, and G. C. Barker, "Biotransformation of tropical lignocellulosic feedstock using the brown rot fungus Serpula lacrymans," *Waste and Biomass Valorization*, vol. 11, pp. 2689-2700, 2020. https://doi.org/10.1007/s12649-019-00581-5