IJAAR PUBLISHING®
Scholarly Journal of Science and Technology Research & Development | ISSN: 2955-0807
Vol. 4, Issue 2 (2025) | www.ijaar.org/sjstrd

Substrate-Dependent Nutritional Variation in *Hypsizygus ulmarius* and *Pleurotus pulmonarius*: A Study on Rice Straw and Groundnut Shell Cultivation

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ABSTRACT

This study investigated the morphological characteristics, Proximate and Vitamin composition of Pleurotus pulmonarius (Fries.) Quel and Hypsizygus ulmarius (Bull.) cultivated on rice (Oryza sativa) straw and groundnut (Arachis hypogeae) shell and their combinations as well as the comparism of the proximate and vitamin contents of both mushrooms were evaluated. Pure mycelium culture of P. pulmonarius and H. ulmarius was aseptically multiplied using sorghum grains. Fully colonized spawn was used to inoculate the substrates which were incubated in the dark room of a mushroom house at 27± 2. Results showed that the substrates supported fructification. Data were collected from fruit bodies harvested from the different substrates and their combinations. Data were subjected to analysis of variance (ANOVA). The morphological characteristics such as pileus diameter, stipe length, fruit body number and weight of the fruit bodies were measured. The highest mushroom fresh weight and dry weight of P. pulmonarius fruit bodies (139.3g and 11.67g) respectively was obtained from Rice straw 100% and that of H. ulmarius (122g and 8g) was obtained from substrate combination Rice Straw 50%+ Groundnut Shell 50%. The proximate analysis showed that the treatment Groundnut shell 100% on the both mushrooms P. pulmonarius and H. ulmarius had the highest contents of protein, fiber, crude fats and moisture. Although, the highest value of carbohydrate, ash content and dry matter was recorded from Rice straw 100% substrates for both mushrooms. Substrate mixtures recorded the highest energy contents for P. pulmonarius (280.39±0.36%) and H. ulmarius (271.31±0.12%). The vitamins assessed were Vitamins A, B_2, B_3, C, D and E. the substrates Groundnut shell 100% had the highest contents of these vitamins from both mushrooms. This study also revealed that the fruiting bodies of P. pulmonarius and H. ulmarius harvested from Groundnut shell 100% substrate holds tremendous promise of nutritious food source, although the mushroom P. pulmonarius on Groundnut shell substrate is of greater economic

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and nutritional value. Therefore, the usage of rice straw and groundnut shell as a medium for mushroom cultivation will assist in reducing environmental pollution caused by the high amount of agricultural wastes.

Keywords: Proximate composition, vitamins, *Hypsizygus ulmarius, Pleurotus pulmonarius*, Rice

INTRODUCTION

Food security exists when all people have physical, economic and social access at all times to safe, sufficient and nutritious food that fulfills their diet requirements for a healthy and active life (WHO, 2012). The Sustainable Development Goals (SDGs) of the United Nations require governments worldwide to end hunger, attain food security, and upgrade nutrition, particularly for the poor and vulnerable. Current food systems produce large quantities of food, but not enough of the required nutrient-rich, plant-based foods needed for healthier and sustainable diets (Siegel *et al.*, 2014; Willett, 2019). Furthermore, significant food losses, particularly due to post-harvest spoilage by microorganisms, exacerbate these food security challenges (Agu *et al.*, 2014; Agu *et al.*, 2015; Agu*et al.*, 2016a; Agu *et al.*, 2016b; Frank & Kingsley, 2014a; Frank & Kingsley, 2014b; Okigbo *et al.*, 2015). Africans have suffered from food insufficiency and malnutrition for decades, largely due to the consumption of staple foods with low micronutrient and protein content. This calls for the production of home-based, protein-enriched foods such as mushrooms that compete as protein and mineral sources among people in developing and underdeveloped countries—an unconventional food fortification strategy (Ishara *et al.*, 2018). The economic and social position of farmers can also be improved through the integration of this non-conventional crop into subsisting agricultural systems.

Mushrooms are fleshy, spore-bearing fruiting bodies of fungi, typically produced above the ground on their food source. All mushrooms are fungi. A mushroom is a macrofungus with a distinctive fruiting body large enough to be seen with the naked eye and picked by hand (Chang *et al.*, 2004). Mushrooms are devoid of leaves and chlorophyll-containing tissues, and therefore are incapable of photosynthetic food production, relying instead on organic matter synthesized by green plants, including agricultural crop residues (Yang *et al.*, 2001). Mushrooms belong primarily to the Basidiomycetes and a few divisions of Ascomycetes, continuing their life cycle through three stages: vegetative growth, reproductive growth, and spore production via fruiting bodies. The cell walls of mushrooms are composed of complex polysaccharides such as glucan and chitin, which provide structural integrity. Mushrooms digest their food externally by secreting enzymes that break down biomass into simpler compounds, which are then absorbed (Chang *et al.*, 2006).

Mushrooms are broadly classified into edible, medicinal, and poisonous types. Major species of edible mushrooms include the button mushroom (*Agaricus bisporus*), oyster mushrooms (*Pleurotus* spp.), paddy straw mushrooms (*Volvariella volvacea*), and shiitake (*Lentinus edodes*). Oyster mushrooms are the second most widely cultivated mushrooms worldwide, after *Agaricus bisporus* (Sanchez, 2010; Kues & Liu, 2000). However, Obodai *et al.* (2003) reported that oyster mushrooms are the third most commercially produced mushrooms in the world market. *Pleurotus* species are popular and widely cultivated in Asia, America, and Europe due to their simple and low-cost production technology (Mane *et al.*, 2007).

Pleurotus pulmonarius, commonly known as the Indian oyster, phoenix mushroom, or lung oyster, is an edible mushroom similar to *Pleurotus ostreatus* (the pearl oyster), but with a few differences. The caps of *P. pulmonarius* are paler and smaller than *P. ostreatus*, and they develop more prominent stems. *P. pulmonarius* also prefers warmer weather and tends to appear later in the summer. Otherwise, the taste and cultivation methods of both species are largely similar (Stamets, 2000). *P. pulmonarius* has a fleshy texture and a pleasant, non-anise-like aroma (Lechner *et al.*, 2004). It has a strong reputation as an edible mushroom, used for centuries in cooking and flavoring due to its desirable taste and unique aroma (Zhang *et al.*, 2013). The cap is fan-shaped, beginning convex and becoming flat or slightly depressed with age. *P. pulmonarius* is the most cultivated oyster mushroom in Europe and North America and is highly marketable in Southeast Asia, Japan, and other regions—though it is more expensive than other mushrooms.

Recent studies have confirmed that *P. pulmonarius* possesses pharmacological properties such as antioxidant, antitumor, anti-inflammatory, immuno-modulating, anti-cholinesterase activities, and potential to reduce vascular complications and blood sugar levels (Nguyen *et al.*, 2016; Zhang *et al.*, 2016; Chioza*et al.*, 2014).

Hypsizygusulmarius, commonly known as the elm oyster mushroom, is an edible fungus. "Hypsi" means “high” and "zygus" means “a yoke,” referring to the mushroom’s typical position high on trees. "Ulm" refers to “elm,” indicating a common substrate. While often confused with oyster mushrooms in the *Pleurotus* genus, it can be distinguished by its gills, which are either non-decurrent or not fully decurrent. The mushrooms appear white to cream in color and are uniform throughout. The stipe extends perpendicularly from the host tree and bends to form the cap, which is usually 6–15 cm in diameter. The stipe lacks rings and connects centrally to the cap.

H. ulmarius is widely cultivated in Asia and Europe due to its high biological efficiency and low-cost production technology (Kumar *et al.*, 2019). It is appreciated for its excellent taste, attractive shape, and large, white fruiting bodies (Baghel *et al.*, 2019). It grows in temperate forests across North America, Europe, and Asia (Stamets, 2015). Although less common than *Pleurotus* species, it thrives on similar substrates such as straw and logs. Although tougher and less flavorful than *Pleurotus* mushrooms, it is still collected in the wild. Compounds in its mycelium and fruiting body have been found to exhibit antioxidant, antitumor, and antidiabetic properties (Greeshma*et al.*, 2016). It is considered a medicinal mushroom in China (Wu *et al.*, 2019).

The aim of this study is to cultivate the fruit bodies of mushrooms and assess the nutritional and vitamin content of *Hypsizygus ulmarius* and *Pleurotus pulmonarius* grown on rice straw, groundnut shell, and a mixture of both substrates.

Materials and Methods

Study Location

The study was conducted in the mushroom house of the Department of Plant Science and Biotechnology, Micheal Okpara University of Agriculture. Umudike, Abia State, Nigeria, which is located between Latitude 7 and 7/05E and Latitude 7 and 5/25N; with humid tropical climate.

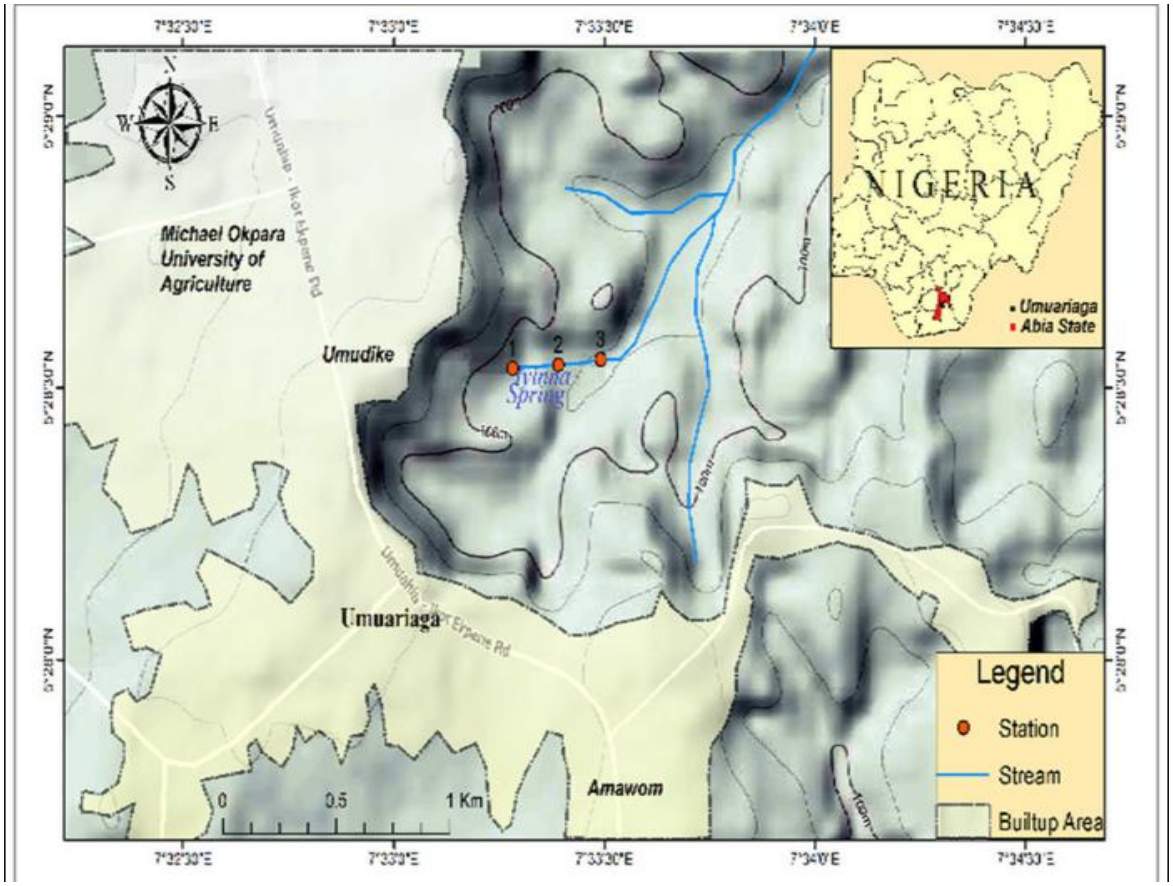


Figure 1: Map of Micheal Okpara University of Agriculture. Umudike, Abia State, Nigeria

Spawn Preparation

The spawn used for the research was prepared in the Science Laboratory of the Department of Plant Science and Biotechnology, Micheal Okpara University of Agriculture. Umudike, Abia State. Spawn of *H. ulmarius* and *P. pulmonarius* was prepared using sorghum grains. The sorghum grains were washed with tap water and soaked over-night. The grains were parboiled for 15-20 minutes using gas cooker as a source of heat for *P. pulmonrius* and *H. ulmarius*. The grains were completely drained of water and spread on a clean mat to drain excess water. The grains were then mixed with 4% (w/w) CaCO3 and 2% (w/w) CaSO4 for *P. pulmonarius* while 2% (w/w) Arsenic and 4% Zinc for *H. ulmarius* to optimize PH and to avoid clumping of the grains (Muhammad, *et al*, 2007). They were subsequently packed in a heat resistant transparent bottle, tightly sealed with Aluminum foil and sterilized in an autoclave at 121°C for 30 minutes. After sterilization, the bottles were allowed to cool before the actively growing pure mycelia of *H. ulmarius* and that of *P. pulmonarius* was aseptically inoculated in their different grains. The bottles containing grains of actively growing mycelium of *H. ulmarius* and also the bottles containing grains of actively growing mycelium of *P. pulmonarius* being inoculated was transferred into the humidity chamber where they were incubated in the dark at 27±2C until the grains were fully colonized by the mycelia (Shyam, *et al.* 2010).

Substrates Preparation

The mushroom house was thoroughly cleaned and washed and the equipment used was disinfected. The buckets used were thoroughly washed and cleansed with antiseptics and they were perforated to ensure aeration. The dried substrate, rice straw was obtained from a rice farmland in Bende, Abia state while the substrate, groundnut shells was obtained from an agricultural farm in Umudike. The rice straw and groundnut shells were chopped into 2-4cm and soaked in water overnight, according to the method of Sharma (2003). The substrates were drained of excess water before transferring in jute bag and then placed into a metallic drum for pasteurization at 100°C for 2 hours. After pasteurization, substrates were allowed to cool over night following the method of Okwulehie *et al* (2008). Pasteurized substrates were brought out of the metallic drum and weighed into 2.5 liter transparent plastic bucket perforated randomly from bottom to the top. The 2.5 liter transparent plastic buckets used were in replicate of 3 which were grouped according to their substrate type and mushroom species.

Spawn Inoculation

The buckets containing 2kg of substrates was inoculated with spawn of *P. pulmonarius* and spawn of *H. ulmarius.* This was done by continuous placing of the spawn between layers of the substrates. After inoculation, the buckets were then covered and transferred to the cropping room where they were placed in wooden racks and covered with black polyethylene sheets from incubation. After eight days (when the substrates were fully colonized by the mycelium), the polyethylene sheets were removed. The humidity of the room was maintained by flooding the room prior to primordial initiation.Fruiting of the mushroom commenced on the different concentration of the substrates from the tenth day after inoculation and were harvested as they matured.



Plate 1: Mushroon Cultivation Chamber

Determination of Proximate Composition of the Mushrooms

Mushroom samples were arranged according to their source of collection and oven-dried at 80°C for 8 hours. The dried mushroom samples were grinded into fine powdery form using manual grinding machine and stored in air tight bottles for further analysis, according to the method of Okwulehie and Odunze (2004). The determination of the proximate composition of the samples followed the procedure of the Association of Official Analytical Chemists (AOAC. 2000). Moisture content was determined by the direct oven drying method. The weight loss after oven drying of each sample (5g) at 105°C to constant weight was expressed as % moisture content (AOAC. 2000). Crude fibre was determined according to the method of Weende. Approximately (5g) of each sample was defatted (during fat analysis). The defatted sample was treated with 200ml of 1.25% of H_2SO_4 solution and boiled under reflux for 30 minutes. The resultant mixture was filtered by washing with several portions of hot water using two-fold muslin cloth to trap the particles. The washed samples was carefully scrapped into a weighed porcelain crucible and dried in the oven at 105°C for 3 hours cooled in a desiccator and weighed. Ash content of 1 g powdered sample was determined as the residue of incineration at 550°C in a muffle furnace (AOAC, 1990). The carbohydrate content of the sample was determined by estimation using the arithmetic difference method described by James (1995). The determination of crude protein was done following the method of Kjedahl (2011). In this method, Nitrogen content was determined and then multiplied with the factor 6.25 to obtain the protein content**.** The fat content was determined using the soxhlet continuous extraction gravimetric method. Vitamin constituents (Vitamin B_2, B_3, C, D, E, A) were determined by atomic absorption spectrophotometry (AOAC, 2005). All proximate analyses of the mushroom powder were carried out in replicate and reported in percentage.

Data Analysis

Percentages and statistical mean were employed to analyze the data generated. The morphological characteristics, proximate and vitamin constituents of *P. pulmonarius* and *H. ulmarius* was analysed using ANOVA.

Results

Table 1: The effect of rice straw and groundnut shell on the morphological characteristics of *Pleurotus pulmonarius*

TREATMENT	S.L	P.D	FBN	FW	DW
RS100	3.28±0.09^{a}	5.18±0.35^{b}	51.67±2.52^{a}	139.33±8.62^{a}	11.67±1.16^{a}
GS100	2.89±0.62^{ab}	3.07±0.49^{ab}	26.67±3.06^{c}	54.67±5.03^{b}	5.00±0.00^{b}
RS 50+GS 50	2.54±0.43^{a}	3.49±0.07^{a}	50.33±11.15^{a}	77.00±24.27^{b}	6.00±1.73^{b}
GS 60+ RS 40	2.67±0.11^{ab}	3.38±0.16^{b}	31.67±3.51^{c}	69.00±8.72^{b}	5.67±1.16^{b}

Data = Mean± standard error, n=5, Values on same row with different alphabets are significantly different at $p<0.05$ levels, P.D= Pileus Diameter, S.L= Stipe Length, FBN= Fruiting Body Number, FW= Fresh Weight, DW= Dry Weight

GS100: Groundnut Shell 100%, RS100: Rice Straw 100%, RS50+GS50: Rice Straw 50%+ Groundnut Shell 50%, GS60+RS40: Groundnut Shell 60%+ Rice Straw 40%

The effect of the substrates on the morphological characteristics of *P. pulmonarius* is presented in Table 1. The result shows that the highest and lowest stipe length value (3.28mm) to (2.54mm) was obtained from *P. pulmonarius* grown on 100% Rice Straw and Rice Straw 50%+ Groundnut Shell 50% substrate respectively The pileus diameter value of *P. pulmonarius* grown on 100% Rice Straw substrate was significantly higher than other experimental substrates. The fruiting body number of *P. pulmonarius* was found to be within the range of (51.67±2.52) to (26.67±3.06) with the highest fruiting body number obtained from the mushroom cultivated on 100% Rice Straw (51.67 ± 2.52^{a}), followed by those grown on Rice Straw 50%+ Groundnut Shell 50% substrate (50.33 ± 11.15^{a}), followed by Groundnut Shell 60%+ Rice Straw 40% (31.67 ± 3.51^{c}) and the least value was obtained from 100% Groundnut Shell (26.67 ± 3.06^{c}). The fresh and dry weight values of *P. pulmonarius* fruit-bodies (139.3g) to (11.67g) respectively was obtained from100% Rice Straw substrate and the least from 100% Groundnut Shell (54.67g and 5.00g)

Table 2: The effect of rice straw and groundnut shell on morphological characteristics of *Hypsizygus ulmarius*

TREATMENT	S.L	P.D	FBN	FW	DW
RS100	2.05 ± 0.07^{ab}	4.17 ± 1.13^{b}	10.67 ± 1.16^{a}	31.67 ± 12.50^{c}	1.67 ± 1.16^{c}
GS100	1.53 ± 0.31^{c}	5.18 ± 0.35^{b}	12.67 ± 0.58^{b}	70.33 ± 3.06^{b}	4.67 ± 0.58^{b}
RS 50+GS 50	1.84 ± 0.23^{bc}	7.76 ± 0.56^{a}	15.67 ± 2.52^{b}	122.00 ± 2.00^{a}	8.00 ± 0.00^{a}
RS 60+ GS 40	2.22 ± 0.11^{a}	8.55 ± 2.23^{a}	23.33 ± 7.10^{a}	140.33 ± 23.46^{a}	8.33 ± 1.53^{a}

Data = Mean± standard error, n=5, Values on same row with different alphabets are significantly different at $p<0.05$ levels, P.D= Pileus Diameter, S.L= Stipe Length, FBN= Fruiting Body Number, FW= Fresh Weight, DW= Dry Weight

GS100: Groundnut Shell 100%, RS100: Rice Straw 100%, RS50+GS50: Rice Straw 50%+ Groundnut Shell 50%, RS60+GS40: Rice Straw 60%+ Groundnut Shell 40%

The result of the effect of rice straw and groundnut shell substrates on the morphological characteristics of *Hypsizygus ulmarius* is presented in Table 2. The result shows that the highest pileus diameter and the highest stipe length value (8.55mm and 2.22mm respectively) was obtained from *H.ulmarius* fruit-bodies grown on Rice Straw 60%+ Groundnut Shell 40%. The least pileus diameter and stipe length was recorded from *H. ulmarius* fruit-bodies from Groundnut Shell 100%. The fruiting body number of *H. ulmarius* obtained ranges from (23.33±7.10) to (10.67 ± 1.16^{a}) with the highest fruiting body number recorded from Rice Straw 60%+ Groundnut Shell 40% substrate and the lowest fruiting body number was obtained from 100% Groundnut Shell substrate. The fresh weight of the mushroom *H. ulmarius* grown on different substrate types was significantly different (Table 4.3). The highest fresh and dry weight of *H. ulmarius* fruit-bodies (140.3g and 8.33g) respectively was obtained from Rice Straw 60%+ Groundnut Shell 40% substrate. The lowest fresh weight was obtained from 100% Groundnut Shell substrate.

Table 3: Comparism of the Proximate Composition of *Pleurotus pulmonarius* and *Hypsizygus ulmarius* cultivated on different substrates

Mushroom	Substrates	Moisture	DM	ASH	FIBER	FAT	CRUDE PROTEIN	CHO	ENERGY
Pleurotus pulmonarius	RS100	4.89±0.02^{d}	95.13±0.04^{a}	14.55±0.35^{a}	1.22±0.13^{d}	13.27±0.17^{d}	24.09±0.36^{d}	42.24±0.32^{a}	275.64±1.57^{c}
	GS100	5.41±0.04^{a}	94.59±0.04^{d}	10.72±0.24^{e}	2.40±0.09^{a}	17.44±0.31^{a}	30.34±0.44^{a}	33.69±0.39^{d}	277.71±0.71^{c}
	RS50+GS50	5.09±0.02^{c}	94.91±0.02^{b}	12.43±0.25^{c}	1.57±0.07^{c}	14.33±0.26^{c}	26.81±0.30^{c}	39.76±0.38^{b}	280.39±0.36^{a}
	GS60+RS40	5.24±0.03^{b}	94.76±0.03^{c}	11.35±0.13^{d}	2.12±0.02^{b}	16.29±0.22^{b}	28.62±0.09^{b}	36.42±0.19^{c}	279.02±0.58^{b}
Hypsizygus ulmarius	RS100	5.41±0.07^{f}	94.59±0.07^{ab}	15.27±0.34^{a}	1.90±0.09^{ef}	15.97±0.12^{g}	20.24±0.27^{f}	41.21±0.23^{ab}	262.91±1.11^{e} ^{f}
	GS100	6.08±0.07^{b}	93.92±0.07^{f}	11.20±0.22^{g}	3.78±0.09^{ab}	19.21±0.13^{b}	26.08±0.45^{a}	33.64±0.25^{f}	272.93±1.48^{a}
	RS50+GS50	5.66±0.07^{cd}	94.34±0.07^{d}	12.63±0.29^{e}	3.21±0.05^{c}	17.90±0.15^{d}	23.26±0.18^{c}	37.32±0.04^{e}	271.31±0.12^{bc}
	RS60+GS40	5.53±0.03^{de}	94.47±0.03^{bc}	13.42±0.38^{d}	2.48±0.07^{de}	16.52±0.25^{f}	22.46±0.27^{d}	39.59±0.83^{b}	270.50±2.91^{d}

Data = Mean± standard error, n=5, Values on same row with different alphabets are significantly different at $p<0.05$ levels.

GS100: Groundnut Shell 100%, RS100: Rice Straw 100%, RS50+GS50: Rice Straw 50%+ Groundnut Shell 50%, RS60+GS40: Rice Straw 60%+ Groundnut Shell 40%, GS60+RS40: Groundnut Shell 60%+ Rice Straw 40%

The comparism of the proximate composition of *Hypsizygus ulmarius* and *Pleurotus pulmonarius* grown on the different substrates is presented in Table 4.5.The results shows that moisture content value of *H. ulmarius* ranges from (6.08±0.07%) to (5.41±0.07%) and that of *Pleurotusp ulmonarius* ranges from(5.41±0.04%) to (4.89±0.02%). The highest moisture content value (6.08±0.07%) was obtained from *H. ulmarius* cultivated on Groundnut Shell 100% which is significantly higher than the moisture content values obtained from *P. pulmonarius*. The highest dry matter content was recorded from the mushroom *P. pulmonarius* grown on Rice Straw 100% (95.13±0.04^{a}). The result shows that the dry matter content obtained from *P. pulmonarius* cultivated on the different substrates was greater than the dry matter obtained from *H. ulmarius* with the lowest dry matter obtained from *H. ulmarius* cultivated on Groundnut Shell 100% (93.92±0.07^{f}). The Elm oyster mushroom (*H. ulmarius)* cultivated on Rice Straw 100% (15.27±0.34^{a}) had the highest ash content which was not significantly different from the ash content value obtained from *P. pulmonarius* cultivated from Rice Straw 100% (14.55±0.35^{a}). The lowest ash content was

recorded from *P. pulmonarius* grown on Groundnut Shell 100%.The crude fiber content obtained from *H. ulmarius* cultivated on Groundnut Shell 100%, Rice Straw 50%+ Groundnut Shell 50% and Rice Straw 60%+ Groundnut Shell 40% was significantly greater than the crude fiber content recorded from *P. pulmonarius* cultivated on the different substrates. The highest fat content was obtained from *H. ulmarius* grown on Groundnut Shell 100% (19.21 ± 0.13^{b}) while the lowest fat content was recorded from *P. pulmonarius* grown on Rice Straw 100% (13.27 ± 0.17^{d}).

Highest crude protein content was recorded from *P. pulmonarius* grown on Groundnut Shell 100% (30.34 ± 0.44^{a}) which was significantly higher than other substrates and the crude. The lowest crude protein content was recorded from *H. ulmarius grown on* Rice Straw 100% (24 ± 0.27^{f}). The highest carbohydrate content was obtained from *P. pulmonarius* (42.24 ± 0.32^{a}) grown on Rice Straw 100% which was significantly higher than other substrates. The carbohydrate content obtained from *P. pulmonarius* grown on Groundnut Shell 100% (33.69 ± 0.39^{d}) was not significantly different from the carbohydrate content obtained from *H. ulmarius* grown on Groundnut Shell 100% (33.64 ± 0.25^{d}).

Substrates mixture gave significantly higher energy value of *P. pulmonarius* fruit-bodies compared to single substrates with the highest energy value obtained (280.39±0.36) from Rice Straw 50%+ Groundnut Shell 50% substrate and the highest energy value of *H. ulmarius* was obtained from Groundnut Shell 100% (272.93 ± 1.48^{a}).

Table 4: Comparism of the Vitamin Composition of *Hypsizygus ulmarius* and *Pleurotus pulmonarius* cultivated on the different substrates.

MUSHROOM	SUBSTRATES	Vit A	Vit B2	B3	C	D	E
Pleurotus pulmonarius	RS100	6.31 ± 0.19^{d}	0.65 ± 0.03^{d}	3.37 ± 0.15^{d}	10.27 ± 1.27^{d}	0.58 ± 0.04^{d}	2.08 ± 0.11^{d}
	GS100	10.69 ± 0.41^{a}	1.18 ± 0.03^{a}	6.28 ± 0.11^{a}	22.73 ± 1.27^{a}	1.03 ± 0.03^{a}	4.00 ± 0.17^{a}
	RS50+GS50	7.27 ± 0.19^{c}	0.76 ± 0.04^{c}	4.46 ± 0.15^{c}	14.67 ± 1.27^{c}	0.76 ± 0.04^{c}	2.51 ± 0.15^{c}
	GS60+RS40	9.30 ± 0.02^{b}	0.96 ± 0.01^{b}	5.44 ± 0.02^{b}	18.53 ± 1.14^{b}	0.86 ± 0.07^{b}	3.16 ± 0.01^{b}
Hypsizygus ulmarius	RS100	4.91 ± 0.18^{gh}	0.39 ± 0.02^{f}	1.90 ± 0.12^{g}	5.87 ± 1.27^{de}	0.44 ± 0.04^{d}	1.10 ± 0.04^{f}
	GS100	9.05 ± 0.32^{ab}	0.71 ± 0.04^{a}	4.57 ± 0.31^{b}	16.87 ± 1.27^{a}	0.95 ± 0.02^{a}	2.85 ± 0.16^{ab}
	RS50+GS50	7.43 ± 0.19^{d}	0.53 ± 0.03^{c}	3.38 ± 0.10^{cd}	13.93 ± 1.27^{ab}	0.78 ± 0.03^{b}	2.19 ± 0.15^{cd}
	RS60+GS40	6.46 ± 0.16^{e}	0.44 ± 0.02^{ef}	2.64 ± 0.24^{f}	10.27 ± 1.27^{cd}	0.65 ± 0.04^{c}	1.70 ± 0.06^{e}

Data = Mean± standard error, n=5, Values on same row with different alphabets are significantly different at $p<0.05$ levels.

GS100: Groundnut Shell 100%

RS100: Rice Straw 100%

RS50+GS50: Rice Straw 50%+ Groundnut Shell 50%

RS60+GS40: Rice Straw 60%+ Groundnut Shell 40%

GS60+RS40: Groundnut Shell 60%+ Rice Straw 40%

The comparative study of the vitamin composition of *Hypsizygus ulmarius* and *Pleurotus pulmonarius* grown on the different substrates is presented in Table 4.9.The results shows that the vitamin A content of *H. ulmarius* ranges from (9.05±0.32%) to (4.91±0.18%) and that of *Pleurotus pulmonarius* ranges from (10.69±0.41%) to (6.31±0.19%). The highest vitamin A content (10.69±0.41%) was obtained from *P. pulmonarius* cultivated on Groundnut Shell 100% which was significantly higher than the vitamin A content obtained from *H. ulmarius*. The highest vitamin B2 content was recorded from the mushroom *P. pulmonarius* grown on Groundnut Shell 100% (1.18±0.03%). The result shows that the vitamin B2 content obtained from *P. pulmonarius* cultivated on the different substrates was greater than the vitamin B2 content obtained from *H. ulmarius* with the lowest vitamin B2 content obtained from *H. ulmarius* cultivated on Rice Straw 100% (0.39±0.02%).

The Indian Oyster mushroom (*P. pulmonarius)* cultivated on Groundnut Shell 100% (6.28±0.11%) had the highest vitamin B3 content. The vitamin B3 content recorded from *P. pulmonarius* grown on the different substrates was significantly greater than the vitamin B3 content obtained from *H. ulmarius*. The lowest vitamin B3 content was recorded from *H. ulmarius* grown on Rice Straw 100%.The vitamin C content obtained from *P. Pulmonarius* cultivated on Groundnut Shell 100% (22.73±1.27%) and Rice Straw 60%+ Groundnut Shell 40% (18.53±1.14%) substrates was significantly greater than the vitamin C content recorded from *H. ulmarius* grown on Groundnut Shell 100% (16.87±1.27%) with the lowest vitamin C content recorded *H. ulmarius* grown on Rice Straw 100%(5.87±1.27%). The highest vitamin D content was obtained from *P. pulmonarius* grown on Groundnut Shell 100% (1.03±0.03%) while the lowest vitamin D content was recorded from *H. ulmarius* grown on Rice Straw 100% (0.44±0.04%). Highest vitamin E content was recorded from *P. pulmonarius* grown on Groundnut Shell 100% (4.00±0.17%) which was significantly higher than vitamin E content obtained from other substrates. The lowest vitamin E content was recorded from *H. ulmarius* grown on Rice Straw 100% (1.10±0.04%).

DISCUSSION

Based on the results of the study, fructification occurred in all the substrates. As regards the features of the mushrooms, substrates types did not influence the colour or the shape of the mushroom harvested. The colours of the fruit-bodies, however, were affected by the stage of the development of the mushroom and the prevailing environmental conditions (Burge, 2008). This result supports the reports of Okwulehie and Okwujiako (2008) that *Pleurotus* species have high saprophytic ability and can grow well in variety of cellulosic substrates.

The results in Tables 1 and 2 showed that there were significant differences in the morphological parameter of *P. pulmonarius* and *H. ulmarius* cultivated on Rice Straw, Groundnut Shell and both substrates mixture. The stipe length value and pileus diameter value of *P. pulmonarius* differed in the different substrates. The highest mean PD and SL was recorded from Rice Straw 100% and it was significantly higher than other substrates. The stipe length value and pileus diameter value of *H. ulmarius* also differed in the different substrates with the highest recorded from Rice Straw 60%+ Groundnut Shell 40% substrate.

The mushroom grown on these substrates used in this study is lower compared to those reported by Okwulehie et al., (2015). For the elm oyster mushroom (*H. ulmarius*), substrates mixture or mixed substrates ensured a smooth transition from vegetative phase to reproductive phase. This agrees with report of Shashirekha and Rajarathnam (2007) who observed that supplementing the coir with rice straw increased the activities of cellulases and protease enzymes from inoculation till the end of fructification. The observation also confirms the work of Kapoor *et al.*, (2009) who noted that the supplementation of different brans (wheat or rice brans) into a substrate for mushroom growth resulted not only in improved linear growth but also in higher activity of cellulases. Fresh weight observed in this study from *P. pulmonarius* ranges from139.3g to54.67g with the highest value obtained from100% Rice Straw substrate. The highest fresh weight of *H. ulmarius* fruit-bodies (140.3g) was obtained from Rice Straw 60%+ Groundnut Shell 40% substrate. From this study, the fresh weight obtained from *H. ulmarius* is greater than the fresh weight recorded *from P. pulmonarius. P. pulmonarius* mushroom cultivated on 100% Rice Straw substrate recorded the highest dry weight value of 11.67g. The highest dry weight value of *H. ulmarius* was recorded from Rice Straw 60%+ Groundnut Shell 40% substrate. Substrate mixture created balanced physical conditions such as good water holding capacity and aeration to support the mycelial growth of *H. ulmarius.*

Results shows that the moisture content obtained from *P. pulmonarius* ranges from(5.41±0.04) to (4.89±0.02) with the highest value recorded from *P. pulmonarius* cultivated on Groundnut Shell 100% as well as the moisture content obtained from *H. ulmarius* with the highest obtained from Groundnut Shell 100%. This result was due to the water holding capacity. The moisture content value of *P. pulmonarius* and *H. ulmarius* mushrooms produced from Rice Straw 100% substrate (5.41%) recorded the same value (Table 3). This might be due to poor water holding capacity of the substrate as compared to other substrates. Wightman (1999) stated that a substrate quality is determined by both the physical characteristics and the chemical characteristics such as high nutrient content. This result was different from the value stated by Mintesnot, *et al*., (2014) who reported 85.6%-93.4% moisture content of *Pleurotussajor-caju*. Saunders *et al*., (2006) noted that a decrease in substrate particle sizes usually result in an increase in water holding capacity. Moisture is necessary for the survival of mushrooms in arid regions.

The Indian Oyster mushroom cultivated on Rice Straw 100% had a dry matter content of (95.13±0.04%) which is significantly higher than the percentage dry matter obtained from other substrates. The Elm Oyster mushroom grown on Rice Straw 100% had a dry matter content of (94.59±0.07). This is in cognizance with the fruiting body number and dry weight obtained from the mushrooms grown on this substrate (Rice Straw 100%). The Ash content value obtained from *P. pulmonarius* cultivated on the different substrates was within the range of 14.55±0.35% to 10.72±0.24% with the highest ash content recorded from the mushroom grown on Rice Straw 100% (14.55±0.35%). The highest ash content value was recorded from *H. ulmarius* grown on Rice Straw 100% (15.27±0.34) which is significantly higher than other ash content value from other substrates. In the both mushrooms (*P. pulmonarius*) and (*H. ulmarius*), substrates mixture resulted in higher percentage ash content compared to the single/pure substrate (Groundnut Shell 100%). Lower percentage ash content of the mushrooms *P. pulmonarius* and *H. ulmarius* grown in the pure substrate (Groundnut Shell 100%) could be as a result of increased percentage composition of other food nutrient like protein content. The percentage crude fiber obtained from *P. pulmonarius* in this study was within the range of 2.40±0.09% to 1.22±0.13% with the highest recorded from Groundnut Shell 100%. Similarly in the mushroom *H. ulmarius*, the highest crude fiber was obtained from the substrate Groundnut Shell 100%. The crude fiber content obtained from *H.*

ulmarius cultivated on these substrates- Groundnut Shell 100%, Rice Straw 50%+ Groundnut Shell 50% and Rice Straw 60%+ Groundnut Shell 40% was significantly higher than the percentage crude fiber recorded from *P. pulmonarius*. The lowest percentage crude fiber obtained from both mushrooms was recorded from Rice Straw 100% substrate. The fat content obtained from *P. pulmonarius* grown on the substrates ranges from (17.44±0.31%)to (13.27±0.17%) with the highest recorded from Groundnut Shell 100%substrate. Also, the fat content obtained from *H. ulmarius* ranges from 19.21±0.13 to 15.97±0.12 with the highest fat content recorded from Groundnut Shell 100%substrate. In this study, fat content obtained from *H. ulmarius* is greater than the fat content obtained from *P. pulmonarius.*

Highest crude protein content was recorded from *H. ulmarius* grown on Groundnut Shell 100% (26.08±0.45) which was significantly higher than other substrates, followed by (23.26±0.18) and (22.46±0.27) recorded from *H. ulmarius* cultivated on Rice Straw 50%+ Groundnut Shell 50% and Rice Straw 60%+ Groundnut Shell 40% substrates respectively. Highest crude protein content was recorded from *P. pulmonarius* grown on Groundnut Shell 100% (30.34±0.44) which was significantly higher than other substrates followed by (28.62±0.09) and (26.81±0.30) recorded from *P. pulmonarius* cultivated on Groundnut Shell 60%+ Rice Straw 40% and Rice Straw 50%+ Groundnut Shell 50% substrates respectively. The difference in the protein content of both mushrooms grown on the substrates could be due to the varying nitrogen content of substrates. The substrate Groundnut Shell 100% contributed to the highest protein content of the fruit-bodies of both mushrooms while the substrate Rice Straw 100% contributed to the lowest protein content. This implies that nitrogen is essential for synthesis of protein in mushroom fruiting bodies. However, the protein content obtained from the mushroom *P. pulmonarius* is significantly greater than the protein content of *H. ulmarius* cultivated on the substrates. Wang, *et al*., (2001) reported that the nature of protein obtained from fruit-bodies is also affected by substrates used. This result also affirms with the finding of Mshandete and Cuff (2007) that protein content of edible mushrooms are influenced by species/strain and the growing substrate. Percentage Carbohydrate content obtained from *P. pulmonarius* grown on Rice Straw 100% (42.24±0.32) was significantly higher than the carbohydrate content from other substrates. Similarly in the mushroom *H. ulmarius*, Rice Straw 100% (41.21±0.23%) substrate recorded the highest carbohydrate content than other substrates. The result from this study indicated that the highest carbohydrate content from *P. pulmonarius* (42.24±0.32%) fruit-bodies is significantly higher than the highest carbohydrate content from *H. ulmarius* (41.21±0.23%). In comparable manner, Moni et al., (2004) obtained carbohydrate content of 40.54% to 47.68% from oyster mushroom; Nuruddin et al., (2010) obtained (33.50%) to (49.58% carbohydrates in *Pleurotus ostreatus* grown on cow dung supplemented rice straw. Substrates mixture gave significantly higher energy value of *P. pulmonarius* fruit-bodies compared to single substrates with the highest energy value obtained (280.39±0.36) from Rice Straw 50%+ Groundnut Shell 50% substrate, followed by (279.02±0.58) from Groundnut Shell 60%+ Rice Straw 40%. But in the mushroom *H. ulmarius*, pure substrate (Groundnut Shell 100% (272.93±1.48) substrate gave the highest energy value than other substrates.

Pleurotus species have been found to have effective vitamin composition in its fruit-bodies. The vitamin constituents of *P. pulmonarius* and *H. ulmarius* as affected by the substrates are shown in Table 4.9. Result shows that the highest Vitamin A (Rectinol) content of *P. pulmonarius* (10.69 ± 0.41^{a}) and *H. ulmarius* (9.05 ± 0.32^{ab}) was obtained from the substrate Groundnut Shell 100%. This result indicated that the vitamin A content of both mushrooms obtained was higher compared to the values obtained by Nwoko, et al., (2017). The highest vitamin B2 (Riboflavin), B3 (Niacin), C (Ascorbic acid), D and E (Tocophenol) content of *P. pulmonarius* and *H. ulmarius* was also recorded from Groundnut Shell 100%. However, in

this study *P. pulmonarius* obtained the highest vitamin constituents value than *H. ulmarius.* In this study, the results shows that substrates used could have significant effect on growth and nutrient composition of the mushrooms. The role of *P. pulmonarius* and *H. ulmarius* in the degradation of these substrates indicates their ability in recycling of agro-waste which was made evident in the research work.

CONCLUSION

This study has shown that Pleurotus species grow fast and this must have proven its choice as one of the most commercially cultivated mushrooms as stated by Husein, et al, (2015). Groundnut shell and Rice straw substrates utilized in this study played a crucial role in the nutrient and vitamin composition of the harvested *P. pulmonarius and H. ulmarius.* As revealed from the result of this study, proximate analysis showed that the mushrooms are a good source of proteins, fibers, fats and oil, carbohydrates and energy. This indicated that the mushrooms are nutritious and can contribute significantly to human health requirements. This study also revealed that the fruiting bodies of *P. pulmonarius* and *H. ulmarius* harvested from Groundnut shell 100% substrate holds tremendous promise of nutritious food source, although the mushroom *P. pulmonarius* on Groundnut shell substrate is of greater economic and nutritional value. Thus, the utilization of these agro-wastes for the production of Oyster mushrooms is more economically practical.

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