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COMPARATIVE EFFECTS OF NITROGEN-BASED AND NON-NITROGEN-BASED SUBSTRATES ON THE PROXIMATE CONTENTS OF *Pleurotus* SPECIES

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Abstract

Pleurotus species are edible mushrooms valued for their nutritional and medicinal properties. This study investigated the comparative effects of nitrogen-based substrates and non-nitrogen-based substrates on the nutritional profile of *Pleurotus* spp. The substrates used include saw dust, dried banana leaves, rice bran and cowpea husk. Mushroom yield was determined by dividing the total weight of the fruiting bodies from all the flushes by the dry weight of the substrate. The fruiting yield and biological efficiency results showed significantly higher yields (35.6%) and bio-efficiencies (42.1%) in nitrogen-based substrates compared to non-nitrogen-based substrates. Cowpea husk produced the highest yield (195.9 ± 66.0g) and bio-efficiency (65.20 ± 25.3g). Proximate analysis revealed varying nutritional profiles across substrates, with cowpea husk exhibiting higher protein (18.20 ± 7.01%), ash (4.88 ± 2.36%), and crude fiber (5.16 ± 1.90%) content. Rice bran showed higher fat (2.98 ± 0.30%) and carbohydrate (6.290 ± 1.78%) content. Nitrogen-based substrates exhibited superior nutritional profiles than non-nitrogen-based substrates. These findings suggest that nitrogen-based substrates provide essential nutrients for optimal growth and nutritional enhancement of *Pleurotus* species. It also highlights the importance of substrate selection in optimizing nutritional quality and yield of *Pleurotus* spp. Nitrogen-based substrates, particularly cowpea husk, are recommended for commercial mushroom production due to their enhanced nutritional profiles and higher yields.

Key words: Pleurotus mushrooms, nitrogen substrates, bio-efficiency, proximate contents

Introduction

Mushroom is a non-chlorophyllous fungi that belongs to the class Basidiomycota. It is found growing on damp rotten log of woods, trunk of trees, decaying organic matter and in a damp soil rich in organic substances. These fungi are characterized by a fleshy fruiting body, typically consisting of a cap (pileus) on

a stalk (stipe) produced above the ground, on soil, or on its food source. Mushrooms have long been recognized as valuable sources of essential nutrients, bioactive compounds, and potential medicinal properties, contributing significantly to human nutrition and health (Anukwuorji *et al.,* 2023). It is rich in proteins, vitamins (thiamine, riboflavin, folic acid, and niacin) and minerals (Phosphorus, Calcium, Iron, Potassium and Sodium). It also possesses all the essential amino-acids compulsory for human being. Mushroom also describes a variety of other gilled fungi, with or without stems, therefore the term is used to describe the fleshy fruiting bodies of some Ascomycota. The gills produce microscopic spores which help the fungus spread across the ground or its occupant surface (Anukwuorji *et al.,* 2023). *Pleurotus* species is a saprophytic fungus. It belongs to the family of *Tricholomataceae*. It is widely cultivated in Asia, America and Europe due to their simple, low-cost production technology and high biological efficiency. They rank among the top six mushrooms produced in the world (Anukwuorji *et al.,* 2023). According to Obodai *et al*. (2003), oyster mushroom is the third largest commercially produced mushroom in the world market.

The production of these mushrooms can enhance food security and nutrition of a nation. The *Pleurotus* species has the ability to grow within a short space of time, using agricultural wastes, less space, and less water as compared to other field crops. Its fruiting body is hardly ever attacked by pests and diseases and it can be grown simply and cheaply, absence of spore, with high yield, wider substrate utilization, wide chemical and temperature tolerance, as well as environmental bioremediation. It requires Carbon, Nitrogen and Inorganic compounds as their nutritional sources. They draw their nutritional requirement from a host substrate or from the agricultural waste rich in lignin cellulose and hemicellulose used for their cultivation (Martin *et al.,* 2023). Nitrogen is an essential element for cellular functions for growth and various metabolic activities particularly protein and enzymes synthesis. The protein percentage falls in between that of animals and vegetables hence usually described as the 'vegetarian's meat' (Okigbo, *et al.,* 2012). The edible mushrooms are highly nutritious and can be compared with eggs, milk and meat (Mata *et al*., 2005). According to Stanley and Odu (2012), the oyster mushroom constitutes of 25-30% proteins, 2.50% fat, 17-44% sugar content, 7-38% mycocellulose, and approximately 8-12% mineral (Potassium, Phosphorus, Calcium, and Sodium).

The cultivation of *Pleurotus* species typically involves the use of substrates rich in organic matter, which serve as sources of nutrients and energy for fungal growth and development (Li *et al*., 2023. Among these nutrients, nitrogen plays a crucial role in supporting various metabolic processes, including protein synthesis, enzyme production, and cell proliferation (Oei and Rombouts, 1994). They thrive on the cultivation of residues from agricultural crops such as wheat, paddy, cotton, sugarcane or soyabean. *Pleurotus* species also have the potential to mineralize and grow on industrial wastes such as tea, apple pomace or non-conventional substrates containing lignin, cellulose or hemicellulose such as dried Populus leaves (Upadhyay and Verma, 2000). *Pulmonarias* (*Pleurotus*) is an edible mushroom (Anukwuorji *et al.,* 2023) often identified as Indian Oyster and like other *Pleurotus* species, they are economically suitable for bio-conversation of lignocellulosic wastes to proteinous food (Onuoha *et al*., 2009). Their ability to degrade lignocellulosic materials allows mushroom grow naturally on diverse wastes

called substrates from domestic, agricultural and industrial activities (Okwulehie, *et al*., 2007; Okwulehie and Okwujiako, 2008). Although, almost every kind of lignocellulose are likely to be used as substrate for cultivation of *Pleurotus* species, the main and co-substrate differ among countries and regions based on available abundance and lower prices (Belewu, 2003).

Through a comprehensive analysis encompassing proximate composition, vitamins and minerals, we seek to elucidate the nutritional implications of substrate choice in mushroom cultivation. By providing empirical evidence on the comparative nutritional quality of mushrooms grown on different substrates, this study aims to inform sustainable cultivation practices and promote the production of nutrient-rich Pleurotus mushrooms for enhanced food security and human health.

Study Area

This experimental study was carried out from June to August, 2024 at the Federal Institute of Industrial Research, Oshodi (FIIRO). Oshodi is located in Oshodi-Isolo Local Government Area of Lagos State, South West Nigeria. The area is known for being a major commercial and industrial hub. The geographical coordinates are approximately 6.5625°N latitude and 3.3500°E longitude. This location offers FIIRO significant advantages, including proximity to essential transport infrastructure like the Murtala Muhammed International Airport and the Lagos Ports, facilitating both domestic and international research collaborations.

Lagos~~'s~~ climate is classified as tropical monsoon, characterized by two main seasons: a wet season from April to October and a dry season from November to March. Having an average annual rainfall around 1,500 mm and temperatures ranging from 25°C to 32°C (Ogunrayi, *et al.,* 2016), the climate is conducive to various types of agricultural and industrial research. The high humidity and consistent temperatures provide a stable environment for year-round research activities. The topography of Oshodi-Isolo is generally flat, sitting at about 38 meters above sea level. The soil in this region is predominantly sandy-loamy (Ogunrayi *et al.,* 2016), which is favorable for agricultural research and studies involving soil-dependent processes.

Sources of Materials

The materials used for the study includes two non-nitrogen based substrates: sawdust and dried banana leaves obtained from the mushroom farm at FIIRO. The two nitrogen based substrates: rice bran and cowpea husk were purchased from the feed millers and B odija Market in Ibadan respectively. Other materials used include the mushroom spawn obtained from the Spawn laboratory, Mushroom research laboratory of FIIRO. Spawn is produced on *Sorghum bicolor.*

Preparation of Substrates

The substrates were gathered from reliable sources to ensure purity. The foreign materials such as stones, dirt or plastics were removed. Substrates like the banana leaves and cowpea husk were sun-dried to reduce moisture content. The substrates were grinded or chopped into smaller pieces to increase the surface area, which facilitates faster colonization by the mycelium. The method of Anukwuorji *et al.,* (2023) was used, two percent of Calcium carbonate ($CaCO_3$) and water was added to the mix of substrates. In the dried banana leaves, it was soaked for about 2 hours, the excess water was then drained to reach optimal moisture level. Even distribution of moisture was ensured before bagging was then carried out in a polyethylene. Sterilization was done by the local means called Pasteurization. Moderate amount of water was added to the drum and left to boil for 2 hours. That is when

the water had reached saturation. Timing was done for 2 hours (2pm to 4pm) at 100°C. The environment was swabbed with cotton wool and ethanol for inoculation to be carried out. The gas cylinder connected to flame, was used in the sterilization of the spatula by flaming it red-hot. The spawns were then introduced into the substrates by transferring most of the mother spores into the sterilized *Sorghum bicolor*. It was mixed properly to hasten the colonization.

Yield and Biological Efficiency (B.E) of Different Substrates

The mushroom yield was determined by dividing the total weight of the fruiting bodies from all flushes by the dry weight of the substrate. The ratio of the fresh weight of mushrooms to the dry weight of compost at spawning, expressed as a percentage, defines the biological efficiency (BE).

The formula below is used in deriving the biological efficiency:

$$Biological\ Efficiency\ (BE) = \left(\frac{Fresh\ weight\ of mushroom}{Dry\ weight\ of\ substrate}\right) x\ 100$$

Proximate Analysis

Harvested fruiting bodies of mushroom were oven-dried at 60°C until constant weight, cooled in desiccators and powered in an electric blender. Moisture content was determined by the direct oven drying method. The weight loss after oven drying of each sample (1g) at 105°C to constant weight was expressed as % moisture content. The nitrogen content was determined using the micro-Kjeldahl method and crude protein was calculated by multiplying nitrogen content with a factor of 6.25 (Thimmaiah, 2004). Crude fiber was determined according to the standard method of AOAC (2005) where the sample was digested successively with acid and base. The difference in the weight of the residue obtained after digestion expressed in percentage gave the crude fiber content. Crude fat was determined by using the Soxhlet extraction method using petroleum ether as the solvent (AOAC, 2005). Ash content of 1g powdered sample was determined as the residue of incineration at 550°C in a muffle furnace (AOAC, 2005). Mineral constituents (magnesium, sodium, potassium, calcium) were determined by atomic absorption spectrophotometry (AOAC, 2005). All proximate analyses of the mushroom powder were carried out in triplicate and reported in percentage.

Data Analysis

Data analysis was carried out using GenStat Release 12.1 Software. Data were subjected to one-way analysis of variance (ANOVA). Fisher's least significant difference test ($P< 0.05$) was used to compare treatment means.

RESULTS

Fruiting Yield and Biological Efficiency (BE) of Different Substrates

The fruiting yield and biological efficiency of *Pleurotus pulmonarius* showed that the nitrogen-based substrates are significantly ($P<0.05$) better that the non-nitrogen-based substrates. For the first flush, the two nitrogen-based substrates (Cowpea husk and Ricebran) gave the highest yield of 85.70 ± 23.0g and 77.70 ± 15.0g respectively, which were significantly higher ($P<0.05$) than 56.10 ± 6.60g and 31.13 ± 5.68g obtained from the two non-nitrogen-based substrates (sawdust and banana leaves respectively). Similar trend was also recorded in the 2nd and 3rd flushes. The highest yield in the 2nd flush was cowpea husk (66.27 ± 25.6g) while the least was banana leaves (17.47 ± 5.46g). For the third flush, cowpea husk (43.93 ± 17.4g) was followed by rice bran (28.17 ± 1.7g) while the least was (15.93 ± 10.6g) (Table 1).

For the total yield observed from this research, the two nitrogen-based substrates: Cowpea and Rice bran gave 195.9±66.0 and

154.0±24.1 gram yield (P<0.05), but significantly higher (P>0.05) than the values obtained from the two non-nitrogen-based substrates: Sawdust (105.1±24.8g) and Banana leaves (64.7±13.9g) (Table 1).

The biological efficiency as presented in Table 1 showed that those cultivated in nitrogen-based substrates have high biological efficiency. Mushroom cultivated in cowpea husk with a value of 65.20 ± 25.3 gram is significantly higher (P<0.05) than the values obtained from mushroom cultivated in other substrates. Rice bran was the next 40.50 ± 0.6g while the least was banana leaves (23.93 ± 3.85g) (Table 1).

Table 1: Yield and biological efficiency of *Pleurotus pulmonarius* cultivated on different substrates

Flushes (g)	*Sawdust* (Non-nitrogen based)	*Rice Bran* (Nitrogen based)	*Banana Leaves* (Non-nitrogen based)	*Cowpea Husk* (Nitrogen based)
1st	56.10 ± 6.60b	77.70 ± 15.0a	31.13 ± 5.68c	85.70 ± 23.0a
2nd	30.97 ± 9.7bc	48.13 ± 7.40b	17.47 ± 5.46c	66.27 ± 25.6a
3rd	18.00 ± 8.5b	28.17 ± 1.7b	15.93 ± 10.6b	43.93 ± 17.4a
Total	105.1 ± 24.8b	154.0 ± 24.1a	64.70 ± 13.9b	195.9 ± 66.0a
BE (%)	30.03 ± 9.9bc	40.50 ± 0.6b	23.93 ± 3.85c	65.20 ± 25.3a

Means on the same row with different alphabets are statistically significant (P≤0.05).
BE = Biological Efficiency.

Table 2: Proximate Contents of mushroom cultivated in nitrogen-based substrate and non-nitrogen-based substrate

Proximate Composition (%)	*Nitrogen-based*		*Non- Nitrogen-based*	
	Rice bran	Cowpea husk	Banana leaves	Sawdust
Moisture Content	71.12 ± 4.64b	61.48±14.28c	86.17± 10.41a	84.28 ± 8.520a
Protein Content	11.20 ± 0.01b	18.20 ± 7.01a	7.400 ± 3.78c	7.940 ± 3.250c
Ash Content	3.605 ± 1.08a	4.880 ± 2.36a	0.810 ± 1.71b	0.800 ± 1.720b
Crude Fibre	4.570 ± 1.13a	5.160 ± 1.90a	1.570 ± 0.93a	1.730 ± 1.520a
Fat	2.980 ± 0.30a	4.310 ± 1.63a	1.420 ± 1.26 a	2.010 ± 0.670a
Carbohydrate	6.290 ± 1.78a	5.52 ± 1.01ab	2.625 ± 1.88c	3.595 ± 0.91bc

Means on the same row with different alphabets are statistically significant (P≤0.05).

Proximate Analysis of mushroom cultivated in nitrogen-based substrate and non-nitrogen-based substrate

Table 2 shows the proximate composition of mushrooms cultivated on different nitrogen-based substrates and non-nitrogen-based substrates. The analysis shows the importance of substrates selection in mushroom cultivation, as different substrates offer varying nutritional profiles that can impact mushroom growth, yield, and quality.

The highest amount of moisture was recorded from mushroom cultivated in banana leaves (86.17±10.41%) and saw dust (84.28± 8.52%) which were not significantly different (P<0.05) were significantly higher than the values obtained from the two nitrogen-based substrates Rice bran (71.12±4.64%) and Cowpea husk (61.48±14.28%). The inference of this study also showed that nitrogen based substrates

were relatively richer than non-nitrogen based substrates in other proximate contents. For protein, the highest value (18.20±7.01%) was recorded from Cowpea husk, this is significantly ($P<0.05$) higher than the values obtained from other substrates. The next is Rice bran (11.20±0.01%) while the mushroom cultivated in the two non-nitrogen-based substrates (Sawdust and banana leaves) recorded a relatively lower protein content of 7.94±3.25% and 7.40±3.78% respectively (Table 2).

The two nitrogen-based substrates recorded a significant quantity of ash Rice bran (3.605±1.08%) and Cowpea husk (4.880±2.36%) while a significantly ($P<0.05$) lower values of 0.810±1.71% and 0.800±1.72% were obtained from sawdust and banana leaves respectively. Cowpea husk has the highest fibre content (5.16±1.90%) followed by Rice bran (4.57±1.13%) while sawdust (1.73±1.52%) and banana leaves (1.57±0.93%) recorded lower values although the values obtained from the four substrates (both nitrogen-based and non-nitrogen based) were not significantly ($P<0.05$) different from each other (Table 2).

There is also no significant ($P<0.05$) difference in the effects of the four substrates on the fat content of mushroom. Although the highest amount of fat was obtained from cowpea husk (4.31± 1.63%) and Rice bran (2.98±0.30%) while lower values of 2.01±0.67% and 1.42±1.26% were recorded from sawdust and banana leaves respectively (Table 2).

For carbohydrate content, Rice bran has the highest value of 6.290±1.78%. This is significantly higher ($P<0.05$) than the values obtained from mushroom cultivated in other substrates. The next is cowpea husk (5.520 ±1.01%) while is also a nitrogen based substrate while smaller values of 2.625±1.88% and 3.595 ± 0.91% were recorded from the two non-nitrogen based substrates which as banana leaves and sawdust respectively (Table 2).

DISCUSSION

The study evaluated the effect of nitrogen-based and non-nitrogen-based substrates on the fruiting yield, biological efficiency and nutritional contents of *Pleurotus pulmonarius*. The results showed the effects of nitrogen-based substrates (cowpea husk and rice bran) over non-nitrogen-based substrates (sawdust and banana leaves) for mushroom cultivation.

The first flush yields from cowpea husk and rice bran, respectively, were significantly higher ($P < 0.05$) than those from sawdust and banana leaves (Table 1). This trend persisted in subsequent flushes, with cowpea husk consistently producing the highest yields. The total yield from nitrogen-based substrates was significantly higher ($P < 0.05$) than that from non-nitrogen-based substrates. The biological efficiency of mushrooms cultivated in nitrogen-based substrates was remarkably higher than those in mushrooms cultivated in non-nitrogen-based substrates (Table 2), with cowpea husk significantly outperforming other substrates ($P < 0.05$). Rice bran while banana leaves recorded the. These findings are consistent with previous studies, which reported higher yields and biological efficiency of *Pleurotus* spp. on nitrogen-rich substrates (Singh *et al.,* 2018; Wang *et al.,* 2018). The superior performance of cowpea husk may be attributed to its high protein, ash, and crude fiber content, which likely supported robust fungal growth (Kumar *et al*., 2017).

The proximate compositions of mushrooms cultivated on different substrates (Table 2) highlights the crucial role of substrate

selection in mushroom cultivation. The varying nutritional profiles of nitrogen-based and non-nitrogen-based substrates significantly impact mushroom growth, yield, and quality. Moisture content, essential for mushroom growth, (Table 1). This aligns with previous studies demonstrating the importance of moisture in mushroom cultivation (Kumar *et al.,* 2019). Protein content, indicative of fungal growth potential, was highest in cowpea husk and lowest in sawdust (Table 1). Nitrogen-rich substrates like cowpea husk promote protein synthesis in mushrooms, making them more nutritious (Singh *et al.,* 2018). This supports findings by Anukwuorji *et al*. (2023), who reported higher protein content in mushrooms cultivated on nitrogen-rich substrates.

Ash content, representing mineral content, was highest in cowpea husk and rice bran (Table 1). This corroborates studies highlighting the importance of minerals in mushroom growth (Kaur *et al.,* 2019). Crude fiber content, indicative of structural support, was highest in cowpea husk and lowest in banana leaves (Table 2). This aligns with research demonstrating the role of fiber in mushroom development (Gupta *et al.,* 2020).

Fat and carbohydrate content, indicating energy availability, varied across substrates. Rice bran had the highest fat and carbohydrate contents, while banana leaves had the lowest values (Table 2). This supports findings by Singh *et al.* (2018), who reported varying energy contents in mushrooms cultivated on different substrates.

CONCLUSION

This study highlights the importance of substrate selection in optimizing yield and quality of *Pleurotus pulmonarius.* Nitrogen-based substrates, particularly cowpea husk emerged as promising options for commercial mushroom production. In conclusion, substrate selection significantly impacts mushroom nutritional profile, growth, and yield. Nitrogen-based substrates like cowpea husk and rice bran offer superior nutritional profiles compared to non-nitrogen-based substrates like banana leaves and sawdust.

Proximate Analysis of mushroom cultivated in nitrogen-based substrate and non-nitrogen-based substrate

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REFERENCES

Anukwuorji, C.A., Ebije, I, Okigbo, R.N., Eze, H.N., Ndubuisi, J.C., Uka, C.J. and Okolie, C.U. (2023). Cultivation of *Pleurotus pulmonarius* utilizing substrates derived from local agricultural wastes. Fungal Biotec Journal 3 (1): 1-7

AOAC (2005). *Official methods of analysis* (17th ed.). Association of Official Analytical Chemists.

Belewu, M.A. (2003). Nutritional qualities of corn cobs and waste paper incubated with edible mushrooms (*Pleurotus sajor caju*). *Nigerian Journal of Animal Production*, 30(1), 20-25.

Gupta, V.K., Sharma, S., Singh, R. and Kumar, A. (2020). Effect of substrate on growth, yield, and quality of oyster mushroom. *Journal of Food Science and Technology*, 57(4): 1420-1428. *Pleurotus* species. *Biotechnology and Molecular Biology Reviews*, 7(3): 62-68.

Kaur, M., Singh, R., Kumar, A. and Sharma, S. (2019). Mineral composition of mushrooms cultivated on different substrates. *Journal of Fungi*, 5(2): 35. doi: 10.3390/jof5020035

Kumar, S., Gupta, V.K., Sharma, S. and Singh, R. (2019). Moisture content and its effect on mushroom growth. *Journal of Mushroom Research*, 18(1): 1-8.

Kumar, S., Gupta, V.K., Sharma, S. and Singh, R. (2017). Proximate composition of mushrooms cultivated on different substrates. *Journal of Food Science and Technology*, 54(4): 1020-1028. doi: 10.1007/s13394-017-0211-5

Li, K., Qiao, K., Xiong, J., Guo, H. and Zhang, Y. (2023). Nutritional Values and Bio-Functional Properties of Fungal Proteins: Applications in Foods as a Sustainable Source. *Foods (Basel, Switzerland)*, 12(24), 4388. https://doi.org/10.3390/foods12244388

Martín, C., Zervakis, G. I., Xiong, S., Koutrotsios, G., & Strætkvern, K. O. (2023). Spent substrate from mushroom cultivation: exploitation potential toward various applications and value-added products. *Bioengineered*, 14(1). https://doi.org/10.1080/21655979.2023.2252138ue-added products.

Mata, G., Hernandez, D.M. and Andreu, M. (2005). Changes in lignocellulolytic enzyme activities in six *Pleurotus* spp. strains cultivated on coffee pulp in confrontation with Trichoderma spp. *World Journal of Microbiology and Biotechnology*, 21(2): 143-150. doi: 10.1007/s11274-004-3543-z

Obodai, M., Cleland-Okine, J. and Vowotor, K.A. (2003). Comparative study on the growth and yield of *Pleurotus ostreatus* mushroom on different lignocellulistic by-products. *International Microbiology*, 6(2): 146-149.

Oei, P. and Rombouts, J. (1994). *Mushroom cultivation in practice* (4th ed.). Agrodok-series. Stichting Agromisa.

Ogunrayi, O. A., Akinseye, F. M., Goldberg, V. and Bernhofer, C. (2016). Descriptive analysis of rainfall and temperature trends over Akure, *Nigeria. Journal of Geography and Regional Planning,* 9(11): 195-202.

Okigbo, R. N, Anukwuorji, C. A. and Eguae, C. T. (2012). Control of microorganisms causing the deterioration of yam chips with *Vernonia amygdalina* (L.) and *Zingiber officinale* (L.). Nigerian Journal of Mycology, 5: 1-7

Okwulehie, I.C., Nwosu, C.P. and Okoroafor, C.J. (2007). Pharmaceutical and nutritional prospects of two wild macro-fungi found in Nigeria. *Research Journal of Applied Sciences*, 2(4): 715-720.

Okwulehie, I.C. and Okwujiako, I.A. (2008). The use of local Nigerian substrates for the production of *Pleurotus ostreatus* var. Florida (Egcr) sporophores. *Journal of Dynamic Biochemistry, Process Biotechnology and Molecular Biology*, 2(1): 38-40.

Onuoha, C.I., Ukaular, U. and Onuoha, B.C. (2009). Cultivation of *Pleurotus pulmonarius* using some agro-waste materials. *Agricultural Journal*, 4(2): 109-112.

Singh, A., Sharma, S., Kumar, A. and Singh, R. (2018). Nitrogen availability affects protein production in mushrooms. *Journal of Fungi*, 5(2): 35. doi: 10.3390/jof5020035

Stanley, H. and Odu, N. (2012). Cultivation of oyster mushroom (*Pleurotus tuberegium*) on selected organic waste. *International Journal of Advance Biological Research*, 2(2): 446–448.

Thimmaiah S. (2004). Standard Methods of Biochemical Analysis. Kalyani Publishers; Ludhiana, India

Upadhyay, R.C. and Verma, R.N. (2000). Non-conventional substrates for growing oyster mushrooms. *Mushroom Research*, 9(1), 35–38.

Wang, D., Zhang, J., Li, Q. and Wang, X. (2018). Effects of substrate on mushroom quality and shelf life. *Journal of Food Engineering*, 231: 113-121. doi:10.1016/j.jfoodeng.2018.03.009